Glyphosate and new cases of resistance - Narrabri

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Keywords

optimising glyphosate performance, annual ryegrass, sowthistle, clethodim, new cases of resistance, NSW random weed survey, germination patterns

GRDC codes

UCS00020, US00084

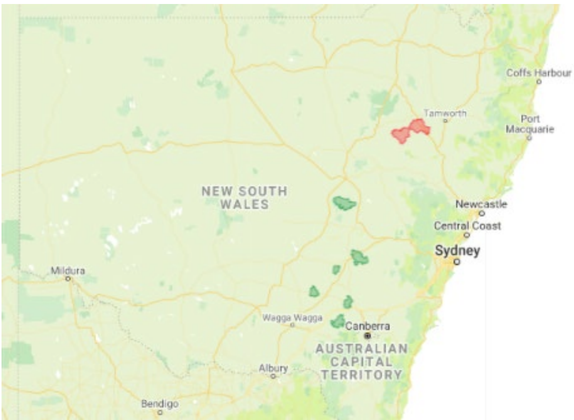
Take home messages

- Increased glyphosate resistant annual ryegrass detected in NSW
- Glyphosate performance can be optimised through attention to application variables such as temperature, plant stress and formulation
- New cases of herbicide resistance have been detected

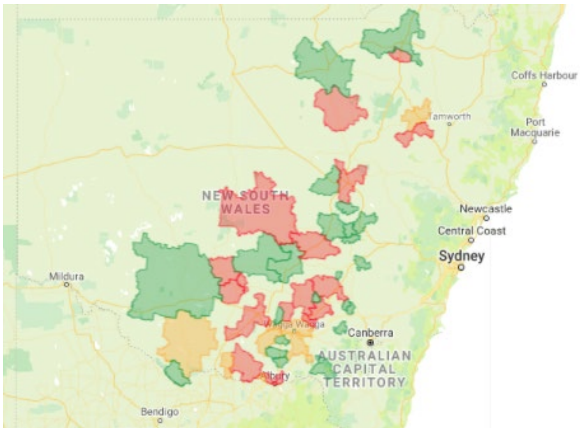
Incidence of glyphosate resistance in NSW

Bayer CropScience provide access to a significant database (Resistance tracker) combining data from national random weed surveys and commercial testing companies (https://www.crop.bayer.com.au/tools/mix-it-up/resistance-tracker). This tracker tool enables the searching of resistance to numerous weed species by postcode and year, with data presented from 2003 (Figure 1).

2003



2009



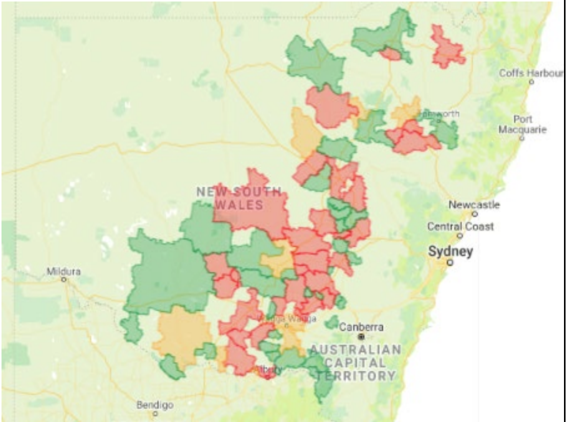
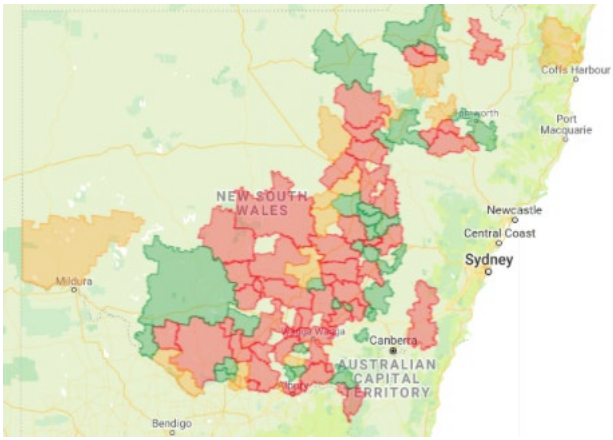
2015	2020
	

Figure 1. Occurrence of glyphosate resistance in annual ryegrass in NSW in 2003, 2009, 2015 and 2020. **Dark green shading** = postcode regions where testing has not detected glyphosate resistance in ryegrass, **orange shading** = postcodes where glyphosate resistance is developing and **red shading** = postcodes where resistance has been detected

2020 season: The early break in 2020 across most southern cropping regions resulted in an opportunity for knockdown weed control. Multiple applications of glyphosate and paraquat were targeted at multiple flushes of weeds, in particular ryegrass, from early autumn to prior to sowing. Plants surviving glyphosate from WA, SA, Vic and NSW were sent to Plant Science Consulting for testing using the Quick-Test method to verify whether herbicide resistance had contributed to survival in the field. The data presented in Figure 2 indicates that 43%, 70% and 79% of ryegrass samples sent from SA, Vic and NSW in 2020 respectively, were confirmed resistant to glyphosate. This highlights that in a majority of cases in NSW, glyphosate resistance has contributed to reduced control in the paddock.

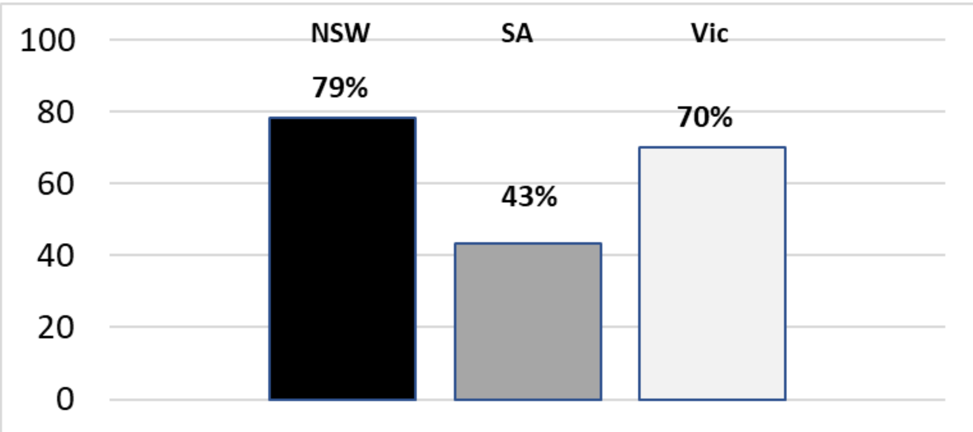


Figure 2. Percent (%) resistance to glyphosate confirmed in farmer ryegrass samples originating from 83 NSW, 37 SA and 74 Vic cropping paddocks treated with glyphosate in autumn 2020. Testing conducted by Plant Science Consulting using the Quick-Test

Discrepancy between resistance testing and paddock failures to glyphosate

In some cases, plants that survived glyphosate in the paddock are not resistant. Reasons for the discrepancy between the paddock and a resistance test can include poor application or application onto stressed plants, incorrect timing, sampling plants that were not exposed to glyphosate, antagonistic tank mixes, inferior glyphosate formulation, poor water quality, incorrect adjuvants, or a combination of the above.

Evolution of glyphosate resistance

Glyphosate was first registered in the 1970's and rapidly became the benchmark herbicide for non-selective weed control. Resistance was not detected until 1996 in annual ryegrass in an orchard in southern NSW (Powles et. al. 1998). Only a few cases of resistance were detected in the following decade (refer to Bayer Resistance Tracker app). The fact that it required decades of repeated use before resistance was confirmed indicated that the natural frequency of glyphosate resistance was initially very low. At the current time there are over a dozen species that have developed resistance to glyphosate in Australia (https://www.croplife.org.au/resources/programs/resistance-management/herbicide-resistant-weeds-list-draft-3/). The most important species in NSW are ryegrass, sowthistle, barnyard grass, fleabane and feathertop Rhodes grass. Ryegrass and sowthistle will be discussed below.

Sowthistle

Differences in the level of control between glyphosate formulations in a glyphosate-resistant sowthistle population from NSW was identified (Figure 3). This finding highlights that significant differences in activity between glyphosate formulations may occur on both susceptible and glyphosate resistant individuals.

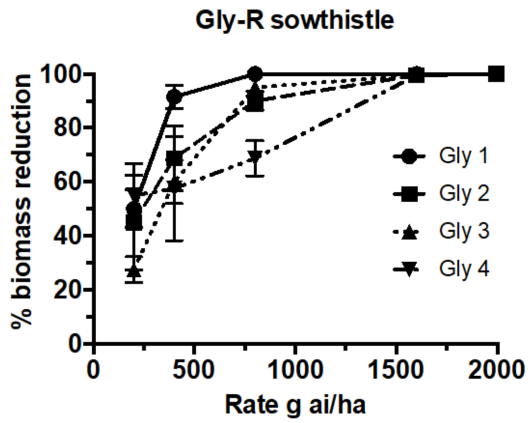


Figure 3. Efficacy of four glyphosate products on control of glyphosate resistant sowthistle as confirmed by outdoor pot trials by Plant Science Consulting

Growth stage and glyphosate rate

Plant growth stage can play an important role in weed control. Even in resistant populations, improved control can often be achieved when targeting younger growth stages. Younger plants tend to have thinner cuticles than older plants improving the speed of uptake. The effect of growth stage and glyphosate rate was investigated in a field trial in NSW on one susceptible and two glyphosate resistant sowthistle populations by Tony Cook, DPI Tamworth (Table 1). Increased control of glyphosate resistant sowthistle was observed at younger growth stages and at higher rates.

Table 1. First cases of confirmed glyphosate resistant sowthistle from Liverpool plains. Data presented as percent biomass reduction at three growth stages. Fallow spray timings from early to late summer. Data courtesy of Tony Cooke, DPI, Tamworth.

Glyphosate rate (g ai/ha)	Growth stage: Early rosette 10cm	Growth stage: Early bolting	Growth stage: Mid-flowering
Susceptible sowthistle- (% biomass reduction)			
360	79	76	0
720	100	81	33
Resistant sowthistle biotype "Yellow" - (% biomass reduction)			
360	55	27	0
720	97	0	0
Resistant sowthistle biotype "CRK" - (% biomass reduction)			
360	64	7	0
720	80	35	5

Application conditions

The effect of temperature on control of glyphosate resistant sowthistle from NSW was recently investigated. Initial trials have confirmed greater control with glyphosate at lower temperatures, particularly of resistant biotypes (Table 2). These findings suggest that applying glyphosate at lower temperatures can improve control of glyphosate resistant sowthistle. At lower temperatures glyphosate remains in liquid form on plant surfaces longer leading to greater uptake, particularly under higher humidity. This allows more hours on the leaf for glyphosate to penetrate. Maximising glyphosate uptake is therefore likely to improve weed control and factors such as lower temperature and higher humidity influence uptake. A pot trial investigating the effect of 20°C and 30°C on glyphosate efficacy identified a higher LD_{50} required at 30°C . This indicates that approximately 2.5x more glyphosate was required to control the resistant populations at 30°C, as opposed to the same populations grown at 20°C

Table 2. Effect of temperature in control of four biotypes of sowthistle from NSW with Glyphosate 540g/L. Data is LD_{50}= dose required to kill 50% of the population.

Biotypes	**Resistance level**	**LD_{50} (g a.i/ha)**	
		20°C	**30°C**
Yellow	strong	439	962
Crocket	strong	389	919
White	weak	132	389
GI	susceptible	135	152

Factors affecting glyphosate performance

There are several contributing factors for the increasing frequency of glyphosate resistance with generally more than one factor responsible. Lower application rates can increase the selection for resistance, particularly in an obligate outcrossing species such as ryegrass, resulting in the accumulation of weak resistance mechanisms to create individuals capable of surviving higher rates. This has been confirmed by Dr Chris Preston where ryegrass hybrids possessing multiple resistance mechanisms were generated by crossing parent plants with different resistance mechanisms.

Other factors that can select for glyphosate resistance by reducing efficacy include:

1. Using low quality glyphosate products and surfactants
2. Mixing glyphosate with too many other active ingredients resulting in antagonism, particularly in low water volumes
3. Using low quality water, particularly hard water. Glyphosate is a weak acid and binds to positive cations (i.e. magnesium, calcium and bicarbonate) that are in high concentration in hard water (i.e. >200 ppm)
4. Applying glyphosate during periods of high temperature and low humidity, resulting in the rapid loss of glyphosate in solution from leaf surfaces and thereby reducing absorption
5. Translocation of glyphosate in stressed plants can be reduced. Maximum glyphosate efficacy relies on translocation to the root and shoot tips. While this occurs readily in small seedlings, in larger plants, glyphosate is required to translocate further to the root and shoot tips to provide high levels of control
6. Shading effects reducing leaf coverage resulting in sub-lethal effects
7. As glyphosate strongly binds to soil particles, application onto dust covered leaves can reduce efficacy
8. Application factors such as speed and nozzle selection, boom height can reduce the amount of glyphosate coverage
9. A combination of the above factors can reduce control thereby increasing the selection for resistance.

Optimising glyphosate performance

The selection of glyphosate resistance can be reduced by considering the points above. A number of important pathways to improve glyphosate performance include:

Avoid applying glyphosate under hot conditions.

A trial spraying ryegrass during the end of a hot period and a following cool change was conducted in October 2019. Ryegrass growing in pots was sprayed at 8am, 1pm and 8pm with temperature and Delta T recorded prior to each application. Control of well hydrated plants ranged between 0% and 40% when glyphosate was applied during hot weather (30 to 32.5°C) and high Delta T (14 to 16.7), with the lowest control when glyphosate was applied at midday (Figure 4). In contrast, glyphosate applied under cool conditions just after a hot spell resulted in significantly greater control (65%-80%), indicating that plants can rapidly recover from temperature stress provided moisture is not limiting, eg. after rainfall.

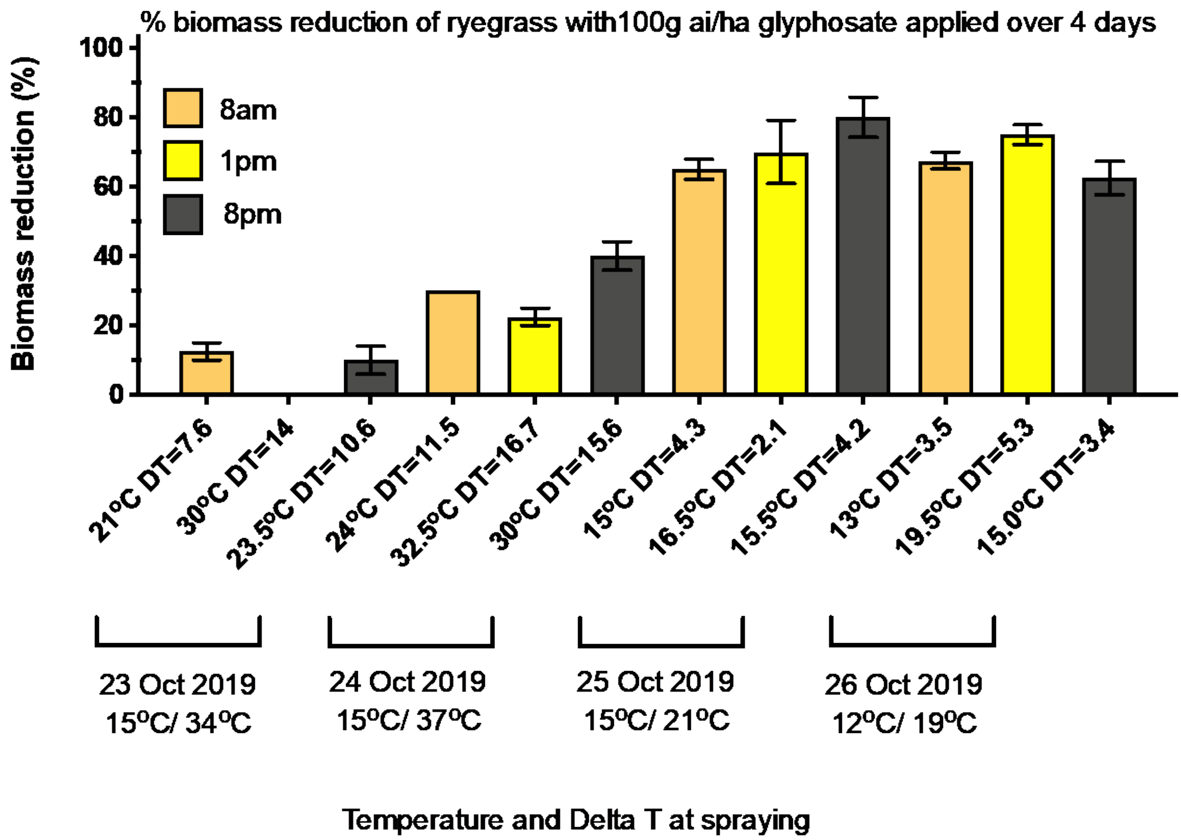


Figure 4. Effect of temperature & Delta T on glyphosate for ryegrass control
(A sub lethal rate was used to differentiate between treatment differences. Plants were grown and sprayed under optimum conditions).

Improving water quality and glyphosate activity by using ammonium sulfate (AMS)

The addition of AMS has several functions. One is to soften water by combining to positively charged ions such as magnesium and calcium common in hard water. The negative charged sulfate ions combine with the positive cations preventing them from interacting with glyphosate and reducing its solubility and leaf penetration. Additionally, AMS has been shown to independently improve glyphosate performance, as the ammonium ions can work with glyphosate to increase cell uptake. In a pot trial conducted with soft water, ammonium sulfate was shown to significantly improve control of ryegrass with 222ml/ha (100g ai/ha) of glyphosate 450 (Figure 5). As a general rule, growers using rainwater (soft) should consider 1% liquid AMS, if using hardwater (i.e. bore, dam) 2% AMS. The addition of a wetter resulted in a further improvement in control.

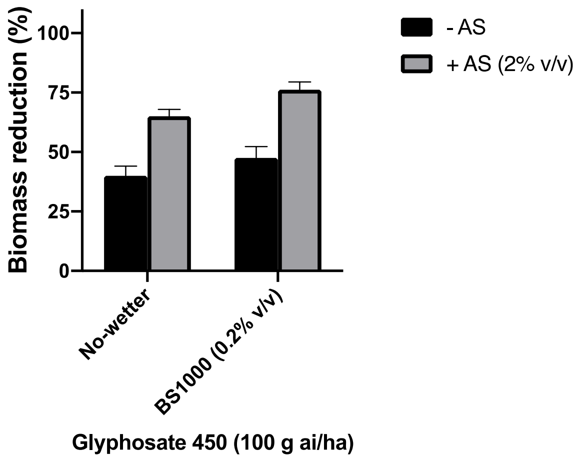


Figure 5. Effect of ammonium sulfate (AS) and wetter on glyphosate for ryegrass control (A sub lethal rate was used to differentiate between treatment differences. Plants were grown and sprayed under optimum conditions).

Herbicide activity can vary at different growth stages

In a pot trial investigating the effect of glyphosate at 4 ryegrass growth stages (1-leaf to 4-tiller), good control was achieved at the 3 older growth stages but not on 1-leaf ryegrass (Figure 6). Most glyphosate labels do not recommend application of glyphosate on 1-leaf ryegrass seedlings because they are still relying on seed reserves for growth. As a consequence, very little glyphosate moves towards the roots.

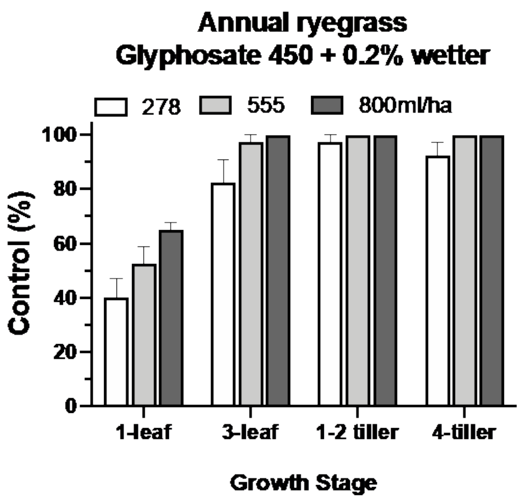


Figure 6. Effect of ryegrass growth stage on glyphosate activity (A sub lethal rate was used to differentiate between treatment differences. Plants were grown and sprayed under optimum conditions).

Double knock

A double knock strategy is defined as the sequential application of two weed control tactics directed at the same weed cohort (germination). The most common double knock strategy is glyphosate followed by paraquat. This has been widely adopted to prevent or combat glyphosate resistance in several weed species, including ryegrass. The first 'knock' with glyphosate controls the majority of the population, with the second 'knock' (paraquat) intended to kill any individuals that have survived glyphosate. Trial work conducted by Dr Christopher Preston (Figure 9) showed that control was optimised when the paraquat was applied 1-5 days after the glyphosate for two glyphosate resistant ryegrass populations. However optimal timing depends on weed size and growing conditions, with at least 3-5 days often being required for full glyphosate uptake and translocation, especially in larger plants. In this study, when the glyphosate resistant plants were left for 7 days before the paraquat application they can stress, resulting in the absorption of less paraquat, reducing control with the second tactic. If growing conditions are poor or plants large, the stress imposed by glyphosate maybe further delayed.

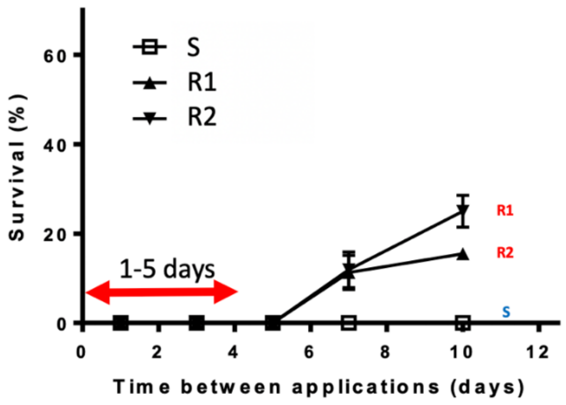


Figure 7. Double knock timing. Glyphosate applied onto a susceptible (S) and two glyphosate resistant ryegrass biotypes (R1 & R2) followed by paraquat 1, 3, 5, 7 and 10 DAA. Trial work conducted by Dr Christopher Preston (The University of Adelaide).

New resistance data

Phalaris

In the 2020 season, 13 phalaris plant samples from populations in paddocks collected from NSW and Qld were tested. Resistance to Group A and/ or B herbicides was confirmed in 10 samples (Table 3).

Table 3. Percent (%) survival of phalaris from NSW and Qld to herbicides. Testing of plants growing in paddocks in 2020 using the Quick test was conducted by Plant Science Consulting. (* herbicide not tested)

Town	**State**	Pinoxaden	Clethodim	Clodinafop	Haloxyfop	imazamox/ imazapyr	mesosulfuron
Widgelli	NSW	*	0	*	*	*	*
Yenda	NSW	*	85	*	*	*	*
Moree	NSW	25	10	40	40	*	0
Moree	NSW	*	*	*	*	*	0
Moree	NSW	65	45	*	50	*	0
Moree	NSW	20	*	*	*	40	50
Goondiwindi	QLD	100	80	100	100	0	0
Bellata	NSW	*	*	*	*	30	60
Croppa Creek	NSW	*	50	*	90	0	*
Pallamallawa	NSW	90	*	*	*	*	*
North Star	NSW	0	0	20	20	60	45
Tulloona	NSW	75	75	*	75	0	15
Tulloona	NSW	0	0	*	0	0	0

Paterson's curse

In the 2019 season, seeds from Paterson's curse that had not been controlled with glyphosate in cotton paddock near Hillston NSW was tested for glyphosate resistance. Survival to glyphosate was confirmed in two trials at 45% survival to 690 g ai/ha at the label rate which controlled a susceptible control population. This is the first reported case of glyphosate resistance in Paterson's curse.

Echinochloa crus-galli

In a 2019 rice crop near Murrambi NSW, poor control of *Echinochloa crus-galli* with Barnstorm® was detected. Seed was sent to Plant Science Consulting for resistance testing in 2020. Strong resistance to Barnstorm (FOP) and lower resistance to Aura® (DIM) was detected, confirming that resistance had contributed to reduced control of *Echinochloa* in the rice field (Figure 8).

This is the first case of Group A resistance in *Echinochloa crus-galli* detected in Australia.

Wild oats

Over 100 samples of wild oats were received in 2020, some as plants during the cropping season and most as seed tests currently being testing. The most common resistance detected is to Group A herbicides. No resistance to triallate has been confirmed. Flamprop-methyl, previously registered as Mataven® and now available as several generic brands, is being included by some growers in their resistant test options.

Ryegrass

The only new case of resistance in annual ryegrass is to paraquat. About half a dozen populations from SA, Vic and WA have been confirmed. Most cases are from situations where paraquat is frequently applied such as lucerne and white clover seed production, fencelines and non-cropped areas. The two Victorian cases however are from cropping paddocks. No cases (to my knowledge) have been detected in NSW. The fact that paraquat resistance has been confirmed in annual ryegrass is of great concern.

Incidence of herbicide resistance in northern region winter annual weeds

Annual ryegrass is the dominant weed of the northern cropping region, despite little or no occurrence of this species in Queensland. Across NSW approximately 70% of annual ryegrass populations are resistant to one or more herbicides (Figure 8), therefore as this species continues to migrate northwards into Queensland it will be resistant populations that establish in these cropping regions.

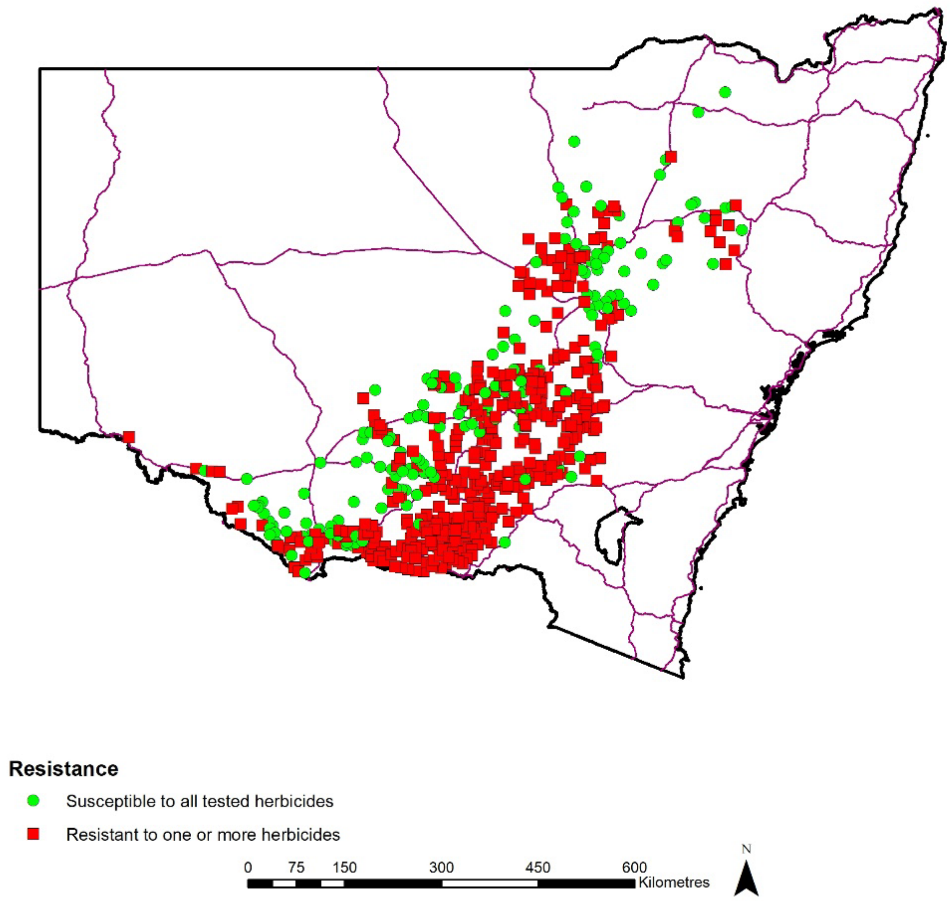


Figure 8. Distribution of northern region annual ryegrass populations that are susceptible (green circles) or resistant (red squares) to one or more herbicides.

The majority of annual ryegrass populations in NSW are resistant to Group A 'fop' and Group B herbicides with some variability between the surveyed regions (Table 4). No populations have been found that are resistant to the newer pre-emergent herbicides, although this has been reported in other states. Of particular concern is the number of populations resistant to glyphosate in some regions.

Table 4. Frequency of herbicide resistance in annual ryegrass populations collected in NSW random surveys (2015 to 2019) (Resistance >20% survival)

	NSW (2015 - 2019)	2015 western NSW	2016 NSW northern	2016 NSW plains	2017 southern NSW	2018 NSW slopes	2019 eastern NSW
	Populations resistant (%)						
diclofop	59	16	32	65	84	77	92
clethodim	2	1	1	1	3	0	12
sulfometuron	50	30	22	35	74	70	82
imazamox/imazapyr	47	8	22	39	75	76	83
trifluralin	1	2	0	0	1	1	2
prosulfocarb + S-metolachlor	0	0	0	0	0	0	0
pyroxasulfone	0	0	0	0	0	0	0
glyphosate	5	6	5	0	7	3	14
Populations screened	608	117	94	111	128	105	53

Wild oat is the next most resistant weed of the northern cropping region with approximately 40% of populations resistant to one or more herbicides (Figure 9). The majority of this resistance is to the Group A herbicide clodinafop (Table 5). Resistance to this herbicide was similar in northern NSW (34%) and Queensland (33%). The highest frequencies of resistance were found in the eastern NSW area where 46% were resistant to clodinafop, with the extent of resistance in Queensland populations higher than all NSW regions except for the eastern area (Table 5).

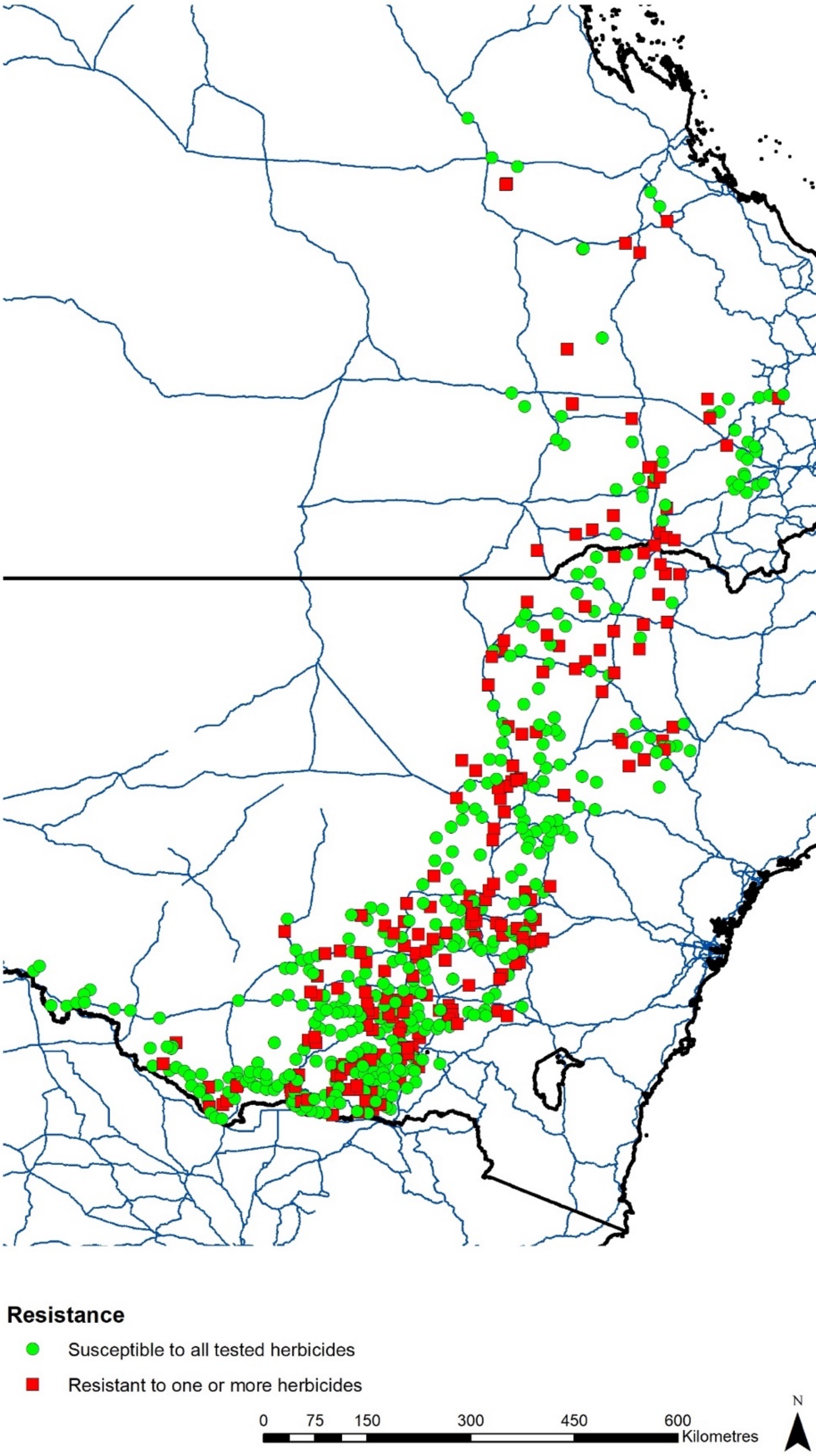


Figure 9. Distribution of northern region wild oat populations that are herbicide susceptible (green circles) or resistant (red squares).

Table 5. Frequency of herbicide resistant wild oat populations randomly collected in recent surveys of the northern cropping region. (Resistant >20% survival) (* = herbicide not screened)

Herbicide	NSW (2015 - 2019)	Qld (2016)	2015 western NSW	2016 NSW northern	2016 NSW plains	2017 southern NSW	2018 NSW slopes	2019 eastern NSW
	Herbicide resistant populations (%)							
Clodinafop	29	33	12	34	31	30	30	46
Clethodim	1	0	0	0	3	0	0	4
Iodosulfuron	4	3	6	4	3	0	0	18
Triallate	0	0	0	0	0	0	0	0
Glyphosate	0	0	*	0	*	0	0	0
Populations screened	511	72	94	122	70	104	93	28

Among the other species collected during the random survey resistance was much lower with no populations of barley grass or brome grass resistant to Group A herbicides and very rare resistance to Group B herbicides. Group F and I resistance was common for wild radish (44% and 29% respectively) and Group B SU resistance in Indian hedge mustard (25%) (Table 6). Of concern is that 3% of barley grass populations were resistant to paraquat (Table 6).

Table 6. Extent of herbicide resistance in populations of the other species collected in random surveys of the northern cropping region (NSW and Qld) (Resistance >20% survival, * herbicide not tested or not applicable for species)

Herbicide group	Barley grass	Brome grass	Wild radish	Indian hedge mustard
	Herbicide resistant populations (%)			
Quizalofop	0	0	*	*
Clethodim	0	0	*	*
Mesosulfuron	1	7	*	*
Chlorsulfuron	*	*	3	25
Imazamox/ Imazapyr	*	2	3	4
Atrazine	*	*	3	0
Diflufenican	*	*	44	6
2,4-D Amine	*	*	29	2
Gramoxone	3	*	*	*
Glyphosate	*	0	0	0
Samples	133	110	33	59

Incidence of herbicide resistance in northern region summer annual weeds

An additional survey collected weed species across northern NSW and Queensland in summer 2016/17. The species collected during this survey included awnless barnyard grass, feathertop

Rhodes grass, fleabane and some additional sow thistle samples. These samples were screened for resistance to glyphosate with a significant percentage of populations for all species except for sow thistle resistant to this herbicide (Table 7).

Table 7: Extent of glyphosate resistance for weed species collected in 2016 summer survey (Includes sow thistle collected in northern NSW and Queensland winter survey)

	Northern NSW		Queensland	
	% Resistant	Populations tested	% Resistant	Populations tested
Awnless barnyard grass	0	5	37	37
Feathertop Rhodes grass	50	2	70	60
Fleabane	100	25	100	36
Sow thistle	7	45	3	62

Summary

Ryegrass blowouts in 2020 were a result of large seedbanks, reduced weed control tactics and herbicide resistance. Using multiple tactics, including new mode of action pre-emergent herbicides, can improve ryegrass control.

While paraquat resistance remains very low, glyphosate resistance in ryegrass is increasing at a rapid rate. The early break in autumn 2020 resulted in the testing of about 200 ryegrass populations prior to sowing with over half confirmed resistant to glyphosate. Decades of strong selection pressure resulting from repeated use coupled with application under suboptimum conditions has played a major role. More efficient use of glyphosate combined with effective IWM strategies is required to reduce further increases in resistance in ryegrass and sowthistle.

Acknowledgements

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