This article is downloaded from

http://researchoutput.csu.edu.au

It is the paper published as:

Authors: Broster, J.C., Koetz, E.A., and Wu, H.
Title: Herbicide resistance levels in annual ryegrass (Lolium rigidum Gaud.) and wild oat (Avena spp.) in southwestern New South Wales
Journal Title: Plant Protection Quarterly
ISSN: 0815-2195
Year: 2013
Volume: 28
Issue: 4
Pages: 126-132
Abstract: In 2010, a random survey was conducted across the cereal cropping zone of southwestern New South Wales to determine the level of herbicide resistance in annual ryegrass (Lolium rigidum Gaud.) and wild oat (Avena spp.) populations. In total, 192 paddocks were visited resulting in 124 annual ryegrass and 104 wild oat seed samples collected for testing. These samples were then screened to the herbicide groups commonly used for annual ryegrass (ACCase, ALS, triazine, dinitroaniline and glycine) and wild oat (ACCase, ALS, thiocarbamate and glycine) control in Australia. The majority of ryegrass samples were resistant to aryloxyphenoxypropionate (56%) and sulfonylurea (53%) herbicides. High levels of resistance were also found for cyclohexanedione (32%) and imidazolinone (38%) herbicides. This represents an increase compared to the 10-14% detected in a 1991 survey. Resistance was found only within ACCase herbicides in the wild oat; 37% were resistant to an aryloxyphenoxypropionate herbicide and 14% were resistant to a cyclohexanedione herbicide. Thirty four percent of the 110 ryegrass samples tested to five herbicide groups (aryloxyphenoxypropionate, cyclohexanedione, ALS, triazine and dinitroaniline) were resistant to two or more herbicide groups, while multiple resistance was only detected in 8% of wild oat samples tested to four herbicide groups (aryloxyphenoxypropionate, cyclohexanedione, ALS and thiocarbamate). Thirty percent of ryegrass and 62% of wild oat samples were susceptible to all herbicides. Resistance levels for both ryegrass and wild oat in this survey were lower than reported for a 2007 survey to the immediate east of this survey. The levels of resistance found for many herbicides, while lower than the adjoining survey, are still significant and highlight the importance of adopting an integrated approach to weed management. This integrated approach is necessary for minimising resistance to commonly used herbicides, and extending their commercial life.

http://researchoutput.csu.edu.au/R/-?func=dbin-jump-full&amp;object_id=53502&amp;local_base=GEN01-CSU01
CRO Number: 53502
Herbicide resistance levels in annual ryegrass (*Lolium rigidum* Gaud.) and wild oat (*Avena* spp.) in south-western New South Wales

J.C. Broster\textsuperscript{A}, E.A. Koetz\textsuperscript{B} and H. Wu\textsuperscript{B}

\textsuperscript{A} Graham Centre for Agricultural Innovation (New South Wales Department of Primary Industries and Charles Sturt University), Charles Sturt University, Locked Bag 588, Wagga Wagga New South Wales 2678, Australia.

\textsuperscript{B} Graham Centre for Agricultural Innovation (New South Wales Department of Primary Industries and Charles Sturt University), Wagga Wagga Agricultural Institute, PMB, Wagga Wagga New South Wales 2650, Australia.

Summary

In 2010, a random survey was conducted across the cereal cropping zone of southwestern New South Wales to determine the level of herbicide resistance in annual ryegrass (*Lolium rigidum* Gaud.) and wild oat (*Avena* spp.) populations. In total, 192 paddocks were visited resulting in 124 annual ryegrass and 104 wild oat seed samples collected for testing. These samples were then screened to the herbicide groups commonly used for annual ryegrass (ACCase, ALS, triazine, dinitroaniline and glycine) and wild oat (ACCase, ALS, thiocarbamate and glycine) control in Australia. The majority of ryegrass samples were resistant to aryloxyphenoxypropionate (56%) and sulfonylurea (53%) herbicides. High levels of resistance were also found for cyclohexanedione (32%) and imidazolinone (38%) herbicides. This represents an increase compared to the 10-14% detected in a 1991 survey. Resistance was found only within ACCase herbicides in the wild oat; 37% were resistant to a
aryloxyphenoxypropionate herbicide and 14% were resistant to a cyclohexanedinone herbicide. Thirty four percent of the 110 ryegrass samples tested to five herbicide groups (aryloxyphenoxypropionate, cyclohexanedione, ALS, triazine and dinitroaniline) were resistant to two or more herbicide groups, while multiple resistance was only detected in 8% of wild oat samples tested to four herbicide groups (aryloxyphenoxypropionate, cyclohexanedione, ALS and thiocarbamate). Thirty percent of ryegrass and 62% of wild oat samples were susceptible to all herbicides. Resistance levels for both ryegrass and wild oat in this survey were lower than reported for a 2007 survey to the immediate east of this survey. The levels of resistance found for many herbicides, while lower than the adjoining survey, are still significant and highlight the importance of adopting an integrated approach to weed management. This integrated approach is necessary for minimising resistance to commonly used herbicides, and extending their commercial life.

Introduction

Herbicide resistant weeds are a major problem in the cropping regions of Australia, with annual ryegrass (Lolium rigidum Gaud.) and wild oat (Avena spp.) being the most important in New South Wales. In the southern cropping region of New South Wales these two weeds were both ranked by agronomists as the first and third most important weeds at that time and for the future (Lemerle et al. 1996). More recently, they were found in over 60% of paddocks in a field survey of cereal crops in southern New South Wales with no other weed species found in more than 23% of paddocks (Broster et al. 2012b).

Annual ryegrass is present in high numbers across the southern Australian cereal zone as a result of its suitability to the climate, extensive use as a pasture species and high seed production (Gill 1996). As a cross-pollinating species, populations of annual ryegrass exhibit
high levels of genetic diversity (Gill 1996) and, while this has assisted in its adaptation to the climatic differences across this region, it also enhances the rate at which herbicide resistance evolves (Gill 1996). Herbicide resistance can potentially be spread by pollen (Busi et al. 2008), however herbicide resistance generally develops in situ through repeated herbicide use.

Wild oat is also prevalent across the southern cropping region of New South Wales. Two species, *Avena fatua* L. and *A. ludoviciana* Durieu, form the majority of in-crop populations with a third species (*A. barbata* Pott ex Link) mainly found in pastures and roadsides (Mansooji et al. 1992). The staggered germination of wild oat places high reliance upon herbicides, both pre-emergent and post-emergent, to control the various cohorts (Jones and Medd 1997). Additionally, as the majority of seed is shed before crop harvest, and non-chemical methods of reducing seed rain to the seed bank are not as effective as for species such as annual ryegrass (Nietschke et al. 1996).

Both annual ryegrass and wild oat are commonly controlled by a number of herbicides, many of which can be used in both the cropping and pasture phases of a rotation. The increased uptake of minimum tillage and stubble retention in many regions has also placed greater emphasis on herbicide use for weed control. Since the release of the first aryloxyphenoxypropionate acetyl coA carboxylase-inhibiting (ACCase) herbicide, diclofop-methyl, in 1978 and the acetolactate synthase-inhibiting (ALS) sulfonylurea herbicide, chlorsulfuron, in 1982, herbicides from these herbicide groups have been extensively used for the control of these species. As a result of the extensive use of herbicides for grass weed control, herbicide resistance developed rapidly in Australia, especially in annual ryegrass (Owen et al. 2007, Broster et al. 2011b, Boutsalis et al. 2012).

While time consuming and expensive, identifying the underlying levels of resistance across large areas through random surveys is important to better understand herbicide
resistance and in designing integrated weed management strategies. Information gained from surveys can aid in the planning of herbicide resistance research and extension for specific areas, as growers with high levels of resistance may have different information needs compared to those who have fewer herbicide resistance concerns (Llewellyn and Powles 2001).

A survey conducted in 1991 established baseline levels for herbicide resistance in much of southern New South Wales (Pratley et al. 1993, Broster et al. 1998). However, as both annual ryegrass and wild oat continue to evolve resistance in the presence of herbicide selection pressure, these surveys need to be conducted regularly. A subsequent survey of much of the same area in 2007 showed marked increases in the level of herbicide resistance in the 16 year period. For ryegrass the incidence of resistance increased from 14% to 81% for diclofop-methyl and from 11% to 70% for chlorsulfuron, while for wild oat the level of resistance to diclofop-methyl increased from 3% to 38% (Broster et al. 2011a, Broster et al. 2011b). Despite the surveys conducted in 1991 and 2007, large areas of the western portion of the southern New South Wales cereal belt have not been surveyed before and there is minimal data on the extent of herbicide resistance available for those areas.

This paper reports the findings of an extensive random survey conducted across the south-western New South Wales cereal belt in 2010 to determine the extent and distribution of resistance in annual ryegrass samples to commonly used herbicides. Differences in the extent of herbicide resistance comparative to a recent survey conducted adjacent to this region are quantified (Broster et al. 2011a, Broster et al. 2011b) as are differences within the surveyed area.

**Materials and Methods**

*Sample collection*
Cropping paddocks in south-western New South Wales were surveyed over a four week period in November and December 2010 prior to harvest. Paddocks were randomly selected at ten kilometre intervals, alternating on the left and right hand side of the survey transects where possible. The location of all sites was recorded using a GPS unit (Garmin 72).

The paddocks were surveyed by two people separately walking an inverted ‘V’ transect, 150 metres into the paddock, and collecting mature seed heads of annual ryegrass and wild oat. Samples were bulked to obtain a single sample of each species for the paddock. A total of 192 paddocks were visited, with 124 annual ryegrass and 104 wild oat seed samples collected for resistance screening. Immediately after collection the seed samples were stored in a glasshouse (10°C minimum, 25°C maximum) until February 2011 when they were threshed and cleaned. At this time the *Avena* species were identified using seed characteristics described by Jacobs and Hasting (1993).

**Resistance screening**

Seeds were sown into either a soil mix (50:50 loam:river wash sand) or a 50:50 peat:sand mix depending upon the herbicide to be screened. For both species all pre-emergent herbicides were sown into the soil mix and, as some ALS herbicides have soil activity, all herbicides in this group were sown in the soil mix for consistency across the herbicide group. The pre-emergent herbicides were all watered within one hour of herbicide application while the post emergent herbicides were not watered until at least one hour after the rainfast period on their label.

**Annual ryegrass** In July 2011 0.2 g (approx. 80 seeds) of seed for each of the 124 samples were planted in plastic punnet trays (330 mm x 280 mm x 60 mm). Each tray contained 14 different samples sown in rows 25 mm apart and then covered to 5 mm in depth.
Three pre-emergent herbicides, chlorsulfuron, simazine and trifluralin (sulfonylurea, trazine and dinitroanoline respectively) were screened. For all three pre-emergent herbicides the seeds were sown in the soil mix. For the chlorsulfuron and simazine treatments, the seeds were sown in rows, covered with 5 mm of the soil mix and the trays then sprayed with herbicide. For the trifluralin treatment, the trays were sprayed, raked to incorporate the herbicide, then seed sown in rows on top of the herbicide and covered with 5 mm of untreated soil.

Samples were screened with five post-emergent herbicides across ACCase, ALS and glycine. Diclofop-methyl, clethodim, glyphosate and imazamox/impazapyr were all applied at growth stage Z12-13 (Zadoks et al. 1974). Seedlings which survived the diclofop-methyl application were sprayed with sethoxydim. As sethoxydim cannot be metabolised by annual ryegrass all resistant populations are the result of changes in the target site (Tardif and Powles 1994). Four weeks after the glyphosate application surviving seedlings were trimmed to 2cm height and sprayed at twice the rate of the first screening to confirm resistance.

Wild oat  In July 2011 approximately 20-30 caryopses from each of the 104 samples were planted in plastic trays (150 mm x 100 mm x 60 mm). The samples were screened with five post-emergent herbicides across ACCase, ALS and glycine. Diclofop-methyl, clethodim, tralkoxydim, mesosulfuron and glyphosate were all applied at growth stage Z12-13 (Zadoks et al. 1974). One pre-emergent herbicide, triallate (Group J) was also screened in this survey.

Seeds were sown in the peat:sand mix for all herbicides except for mesosulfuron and triallate. For the triallate treatment, the trays were sprayed, then raked to incorporate the herbicide and the seeds were sown on top of the herbicide-treated soil and covered with 5 mm of untreated soil.

For both species, trays were kept in a temperature controlled glasshouse (10°C minimum, 25°C maximum) and watered and fertilised as required. Two weeks after sowing,
all samples in the post-emergent herbicide treatments were counted and thinned to a maximum of 20 or 10 plants per sample for the ryegrass and wild oat, respectively. Three replicates were sown for all samples except where seed numbers were limited.

*Herbicide application*

The herbicide resistance testing protocol was adapted from Broster and Pratley (2006), with herbicides only applied at the label recommended rate similar to previous surveys (Broster et al. 2011a, Broster et al. 2011b, Broster et al. 2012a) (Table 1). All herbicides were applied using an automated laboratory-sized cabinet sprayer with a moving boom applying a water volume of 77 L ha⁻¹ equivalent from a flat fan nozzle at 300 kPa pressure. Adjuvants were added to herbicides as per label requirements (Table 1). A standard susceptible biotype (CSU–S1) and known resistant biotype, where available, were included with each cohort of samples. Due to the limited seed availability of some samples, not all of the 124 ryegrass or 104 wild oat samples were screened with all herbicides, or for all replicates.

*(Insert Table 1 here)*

*Herbicide evaluation*

All samples were assessed between 21 and 28 days after treatment. Seedlings in post-emergent treatments were counted before and after treatment to enable survival percentages to be calculated. Samples sprayed pre-emergent were rated visually from 0 (no germination) to 10 (no visual difference from untreated control).

Results were analysed by ANOVA using Genstat version 11.1 (GenStat 2008) and the standard error for each herbicide determined. As the standard error for none of the herbicides screened exceeded 1.5 for the pre-emergent or 10% for the post emergent herbicides standard
thresholds were used as this ensured samples classified as developing resistant or resistant were significantly different from the susceptible control. Samples were classified as resistant if the mean survival percentage for all replicates was greater than 20% for post-emergent herbicides or a visual score of greater than 2.5 for pre-emergent herbicides (Broster and Pratley 2006). Samples with survival percentages of between 10 and 19% for post-emergent herbicides or a visual score of between 1.5 and 2.5 for pre-emergent herbicides were classed as developing resistance. Samples were classed as susceptible if survival was less than 10% or a visual score of below 1.5.

Four regions were determined within the surveyed area approximately based on shire boundaries. The two southern regions (C and D) were combined for the wild oats due to limited sample numbers. When comparing differences between the different regions and other sample cohorts z-tests were used.

Results

Annual ryegrass

Among samples collected for this survey, resistance was common to all of the tested ACCase inhibiting and ALS-inhibiting herbicides. Of the samples screened to diclofop-methyl, 56% (69 samples) were classed as resistant (combined resistance and developing resistance categories) and for tralkoxdim it was 32% (39 samples). For chlorsulfuron and imazamox/imazapyr the proportion of samples classed as resistant was 53% (63 samples) and 38% (44 samples) respectively (Table 2). Upon subsequent screening to sethoxydim, 70% of the diclofop-methyl resistant samples also exhibited resistance to sethoxydim, equal to 39% of samples. Only 4% of the samples were resistant to clethodim and no samples were classed as resistant to simazine, trifluralin or glyphosate (Table 2).
Compared to a survey conducted in 2007 in the area immediately to the east of this survey, the level of herbicide resistance in annual ryegrass for this survey was much lower. In this survey, a lower percentage of samples were classed as resistant to diclofop-methyl (56% cf. 81%; P<0.001), imazamox/imazapyr (38% cf. 65%; P<0.001), chlorsulfuron (53% cf. 70%; P<0.05) and clethodim (4% cf. 21%; P<0.001) (Broster et al. 2011b).

When two or more herbicides within a group were tested, some differences in the resistance status of the samples could be found. Among the cyclohexanedione herbicides, no samples susceptible to either sethoxydim or tralkoxydim were resistant or developing resistant to clethodim. All five samples classed as resistant or developing resistant to clethodim were resistant to both sethoxydim and tralkoxydim with survival levels to both herbicides greater than 40% for all but one sample to sethoxydim (30%). Of the samples tested to both chlorsulfuron and imazamox/imazapyr of these 51 were classed as susceptible to both, 31 were resistant to both, nine were developing resistance to one herbicide and resistant to the other. Among the remaining 26 samples one was resistant and three developing resistance to imazamox/imazapyr but susceptible to chlorsulfuron, while nine were resistant and 13 developing resistance to chlorsulfuron but susceptible to imazamox/imazapyr.

**Wild oat**

Samples of wild oat were classed as resistant to only two of the six herbicides screened, diclofop and tralkoxydim, both ACCase inhibiting herbicides (Table 3). Thirty-eight samples (37%) were resistant to diclofop-methyl and 14 to tralkoxydim (14%). None of the samples screened were found to be resistant to clethodim, mesosulfuron, triallate or glyphosate (Table 3).
Multiple resistance

Of the 124 ryegrass samples, 109 were screened to all five groups of selective herbicides (aryloxyphenoxypropionate, cyclohexanedione, ALS inhibitor, triazine and dinitroaniline). If a sample was resistant or developing resistance to either herbicide in one of the two groups (cyclohexanedione or ALS inhibiting) where two herbicides were tested, it was classified as resistant to that group. Thirty two samples (30%) were susceptible to all groups, 23 (21%) were only resistant to one group, 17 of which were resistant to an ALS inhibiting herbicide and six to an aryloxyphenoxypropionate herbicide. Twenty one (19%) of the samples were resistant to two groups, 11 of these were resistant to aryloxyphenoxypropionate and cyclohexanedione herbicides with the other ten resistant to aryloxyphenoxypropionate and ALS inhibiting herbicides. Thirty three samples (30%) were resistant to three herbicide groups while no samples were resistant to four or more herbicide groups.

Seventy six wild oat samples were screened to four groups of selective herbicides (aryloxyphenoxypropionate, cyclohexanedione, sulfonylurea and thiocarbamate), of these 47 (62%) were susceptible to all herbicide groups, 23 (30%) were resistant or developing resistant to one herbicide group and six samples (8%) were resistant to two herbicide groups.

Within survey differences

Four distinct regions were used to evaluate differences within the surveyed area (Figure 1). Region A (47 samples - west of West Wyalong and north of Narrandera), B (28 samples - east of West Wyalong and north of Wagga Wagga), C (17 samples - south of Narrandera and
north of Jerilderie) and D (32 samples - south of Jerilderie) all had distinctly different resistance profiles.

The percentage of total resistant samples (combined resistant and developing resistance) to diclofop-methyl at 40% was lower in Region A compared with the other three regions where it was between 64 and 66% (P<0.001) (Figure 1a). A significantly higher percentage of the samples resistant to diclofop-methyl in Region D (90%) were also resistant to sethoxydim compared to Regions A (42%; P<0.001) B and C (both 72%; P<0.01) and while both Regions B and C had higher levels of resistance than Region A (P<0.001). Forty seven percent of samples were resistant to tralkoxydim in both Region C and D significantly higher than in Region A (17%; P<0.001) and B (29%; P<0.01). Although the percentage of samples resistant to clethodim was much lower, differences were still apparent between the regions, with 9% resistant in Region D compared with 0% in Region B(P<0.01).

Similar trends between the regions were recorded for both of the ALS inhibiting herbicides screened, chlorsulfuron (Figure 1b) and imazamox/imazapyr. As with the other herbicides Region D had the highest percentage of resistant samples, 87% and 63% for chlorsulfuron and imazamox/imazapyr respectively, higher than all other regions (P<0.001 except Region B imzamox/imazapyr P<0.01). Region B had the next highest level of resistance to the two ALS inhibiting herbicides tested (54% and 43%) significantly higher than both Region A (36%; P<0.05 and 25%; P<0.01) and Region C (31%; P<0.01 and 13%; P<0.001).

In annual ryegrass when comparing the region with the lowest resistance level to the region with the highest found the increases ranged from 50% for diclofop-methyl, 180% for tralkoxydim and chlorsulfuron to a 300% increase in the incidence of resistance to imazamox/imazapyr. For wild oat the incidence of resistance in Region B compared to Region A was 66% and 100% higher for diclofop-methyl and tralkoxydim respectively.
The level of multiple resistance also varied between regions, of the samples tested to the five herbicide groups, a higher percentage of samples from Region D (61%) were resistant to three groups than the other regions (A - 10%, B – 36% and C – 15%; P<0.001) and more from Region B than Regions A and C (P<0.001) Samples from Region A were more likely to be susceptible to all herbicides than the other regions (A - 45% cf. B – 29%; P<0.05, C – 23%; P<0.01 and D – 11%; P<0.001). There were minimal differences between the regions in terms of the percentage of samples resistant to either one or two herbicide groups.

(Insert Figure 1 here)

After species identification of the wild oat, 50% of samples appeared to be *Avena ludoviciana*, 20% *A. fatua* with the remaining 30% of samples containing both species. Samples containing *A. fatua* had higher levels of resistance than those consisting of only *A. ludoviciana* for both diclofop-methyl (P<0.001) and tralkoxydim (P<0.05). The majority of wild oat samples came from Regions A and B (66 and 25 samples respectively) as the seed had been shed prior to sample collection in many crops in Regions C and D limiting sample numbers to five and eight respectively, the data. Having said this, wild oat plants were present in 13 and 16 paddocks in Regions C and D respectively.

There was a higher incidence of resistance in *A. fatua* populations compared to *A. ludoviciana*, however this is potentially biased by the majority of wild oat samples being collected in Region A where both resistance was lowest and *A. ludoviciana* the most common species. Eighty one percent of the samples containing only *A. ludoviciana* were collected from Region A; this was 64% of the samples from that region compared to Region B where 28% of the wild oat samples consisted of only *A. ludoviciana*. Overall, the level of wild oat resistance was lower in Regions C and D compared to Regions A and B.
resistance was significantly lower in Regions A and CD compared to Region B for diclofop-methyl (32% & 31% cf. 52%; P<0.01) but not tralkoxydim (11% cf. 20%; P>0.05).

Discussion

Comparison to other surveys – annual ryegrass

This survey found 56% of annual ryegrass to be resistant to diclofop-methyl compared to 81% in the area of southern New South Wales in 2007 immediately to the east of this survey (Broster et al. 2011b), 68% in Western Australia (Owen et al. 2007) and 76% in central South Australia (Boutsalis et al. 2012). Other surveyed regions that had similar levels of resistance to diclofop-methyl to that found in this survey were the Victorian Wimmera and south-eastern South Australia (Boutsalis et al. 2012), while resistance levels reported here were markedly higher than those found in the Mallee regions of both South Australia and Victoria where resistance was observed in six and twelve percent of populations, respectively (Boutsalis et al. 2012).

Similarly, resistance to chlorsulfuron was lower (53%) than that found in the 2007 New South Wales (70%) and central South Australia (73%) surveys and also lower than the incidence of resistance to sulfometuron (88%) found in Western Australia (Owen et al. 2007, Broster et al. 2011b, Boutsalis et al. 2012).

These differences in herbicide resistance represent the level of selection pressure placed on the ryegrass populations in the various regions. The more intensely cropped regions are likely to have higher levels of herbicide resistance both in the percentage of populations resistant to a given herbicide as well as the number of herbicides to which these populations are resistant.

The area sampled in this survey had not been previously surveyed for herbicide resistance with the exception of much of Region B which had been surveyed in 1991.
Resistance to diclofop-methyl and chlorsulfuron across the entire 1991 survey region was recorded in 14% and 11% of samples respectively (Pratley et al. 1993). In Region B of the 2010 survey the incidence of resistance was 64% and 54% for diclofop-methyl and chlorsulfuron.

The higher proportion of samples susceptible to all herbicides compared to other surveys, especially in region A, again indicates the reduced level of selection pressure for resistance through reduced cropping intensity in comparison to many of the other surveyed regions.

Although this survey reported no ryegrass samples resistant to glyphosate, a semi-targeted survey around Hillston conducted in 2012 found 21% of ryegrass samples to be resistant or developing resistant to glyphosate (Haskins pers comm.) The ability to grow summer crops in some areas within this survey may place additional pressure on the development of resistance to glyphosate. The potential for glyphosate to be used in both summer and winter fallows could result in more applications over a given time period. This would result in increased selection for resistance where a new generation of a species are treated with each subsequent herbicide application. Alternatively, where multiple herbicide applications are applied to a single generation of a species only the most resistant of plants will survive with no susceptible escapes also increasing the selection pressure for resistance. Resistance to glyphosate has occurred in many annual ryegrass populations, as well as in awnless barnyard grass (Echinochloa colona (L.) Link) (Preston 2013). Awnless barnyard grass and other Echinochloa species (eg. Echinochloa crus-galli (L.) Beauv.) are common across the irrigation areas within this survey area (Pratley et al. 2008).
Comparison to other surveys – wild oats

Unlike annual ryegrass, the incidence of diclofop-methyl resistance in wild oat is no different to that of the 2007 survey conducted immediately east of the area covered by this survey (37% cf. 38%) and no resistance was reported for any of the other herbicides common to both surveys (Broster et al. 2011a).

The lower level of resistance in wild oat within this survey compared with ryegrass (Tables 2 and 3), even in paddocks where both species were collected, agrees with the previous survey conducted to the east of this survey (Broster et al. 2011a, Broster et al. 2011b). Many of the herbicides used control both species and therefore the herbicide selection pressure should be similar, meaning the difference in herbicide resistance is due to the characteristics of the species themselves. Wild oat is a self-pollinating hexaploid species while ryegrass is a cross-pollinating diploid species. Yu et al. (2013) stated that the evolution of resistance is more complex in hexaploid wild oat compared with diploid grass species. Other factors such as the self-pollination, seed bank longevity and staggered germination of wild oat, and even lower population numbers may also influence the rate at which resistance develops in wild oat compared with ryegrass.

Within survey differences

While this survey covered a much smaller geographical area than the 2003 Western Australian survey (Owen et al. 2007), or the surveys of South Australia and Victoria conducted between 2005 and 2009 (Boutsalis et al. 2012), there were still marked differences between regions within this survey with respect to the incidence of resistant populations. There were marked differences between the regions with the lowest and the highest incidence of herbicide resistance.
The marked variations between regions within the surveyed areas reflect differences in both the crops grown and the methods used to sow those crops. Region C had the lowest level of resistance to the ALS herbicides, significantly lower than Regions B and D. Compared with these regions a greater proportion of the winter crop sown in this region was canola or grain legumes (Australian Bureau of Statistics 2013). The use of ALS inhibiting herbicides is restricted with these crops (Brooke et al. 2013) thereby reducing their use and limiting the selection pressure for resistance development and subsequent resistance levels to these herbicides in this region.

Region A had the lowest levels of resistance in ryegrass to the ACCase herbicides and lower levels (compared with Regions B and D) to the ALS herbicides. For wild oat the level of resistance to ACCase herbicides was lower compared with Region B, the only two regions from which more than 10 samples were collected. In this region, wheat formed a higher proportion of winter crop grown than the other regions (Australian Bureau of Statistics 2013) providing greater potential for rotation between the ACCase and ALS herbicides (Brooke et al. 2013) reducing the selection pressure on both herbicide groups. A greater percentage of the crop in Region A was sown after two or more cultivations (Australian Bureau of Statistics 2013) further reducing selection pressure by, either using cultivation as a method of weed control, or possibly the use of fallow both controlling weeds and reducing the cropping intensity. However, since these data were obtained (2001 and 2002 seasons) it is likely that the amount of cultivation has been reduced to take full advantage of the lower rainfall experienced in this region placing increased emphasis on herbicides for weed control and increasing selection pressure for resistance.

Irrigated summer crops, such as rice and cotton, form a large part of the farming rotation in Region A, and parts of Region D, and this can also slow the development of herbicide resistance. This is because alternative herbicides can be used in crop, or knockdown
herbicides used in a winter fallow situations thereby reducing selection pressure on the selective herbicides.

As only a portion of surveyed region had previously been surveyed, no general conclusions can be drawn as to the overall rate of resistance change. However, much of Region B had been surveyed in 1991, with the proportion of ryegrass populations resistant to diclofop-methyl and chlorsulfuron increasing over this time from 14% to 64% and 11% to 54% respectively. The annual rate of increase of 2–2.5% was much lower than the 4% recorded both in the area of southern New South Wales surveyed in 2007 (Broster et al. 2011b) and in Western Australia (Llewellyn and Powles 2001, Owen et al. 2007). The lower rate of increase may reflect a combination of both, reduced cropping intensities and inputs over much of the surveyed area, as well as the adoption of integrated weed management practices due to increased awareness of herbicide resistance.

With the high proportion of sites containing both species (44% in this survey), it would be expected that regions with a higher incidence of resistance in the ryegrass populations would also have a higher incidence of resistance in the wild oat populations. If more samples had been collected from the southern part of the survey, where the wild oat seed had shed before collection and the incidence of ryegrass resistance was higher, the incidence of wild oat diclofop-methyl resistance may have been higher in this survey.

**Conclusion**

Significant levels of resistance were present for diclofop-methyl and tralkoxydim (both ryegrass and wild oat) and ALS herbicides (ryegrass only). The low incidence of clethodim resistance in ryegrass and the absence of resistance in ryegrass and wild oat to all other tested herbicides is a positive finding. However, where herbicide resistance is present in some of the available herbicide groups, greater selection pressure for resistance is placed on the
remaining herbicides. Reducing the number of individuals in a weed population treated with herbicides through the use of non chemical weed control methods will prolong the effective life of the herbicides which currently have a high incidence of resistance, while also reducing the pressure on resistance development in the herbicides with lower incidence of resistance.

**Acknowledgements**

This survey was conducted as part of a larger project funded by the Grains Research and Development Corporation. The authors acknowledge the assistance of technical staff from New South Wales Department of Primary Industries and students from Charles Sturt University for their assistance in the resistance screening and Professor Leslie Weston for her suggestions on the manuscript.
References


Table 1. Herbicides and rates used for herbicide resistance screening for annual ryegrass and wild oat in south-western New South Wales.

<table>
<thead>
<tr>
<th>Herbicide</th>
<th>Herbicide group</th>
<th>Pre- or post- treatment</th>
<th>Rate (g ha(^{-1}) a.e.)</th>
<th>Adjuvant (rate % v/v)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Annual ryegrass</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>diclofop-methyl</td>
<td>aryloxyphenoxypropionate</td>
<td>Post</td>
<td>375</td>
<td>Chemwet 1000 (0.25)</td>
</tr>
<tr>
<td>tralkoxydim</td>
<td>cyclohexanedione</td>
<td>Post</td>
<td>172</td>
<td>Supercharge (1.0)</td>
</tr>
<tr>
<td>sethoxydim</td>
<td>cyclohexanedione</td>
<td>Post</td>
<td>186</td>
<td>DC Trate (1.0)</td>
</tr>
<tr>
<td>chlorsulfuron</td>
<td>sulfonylurea</td>
<td>Pre</td>
<td>15</td>
<td>-</td>
</tr>
<tr>
<td>imazamox/imazapyr</td>
<td>imidazolinone</td>
<td>Post</td>
<td>48</td>
<td>Hasten (0.5)</td>
</tr>
<tr>
<td>simazine</td>
<td>triazine</td>
<td>Pre</td>
<td>1260</td>
<td>-</td>
</tr>
<tr>
<td>trifluralin</td>
<td>dinitroaniline</td>
<td>Pre</td>
<td>816</td>
<td>-</td>
</tr>
<tr>
<td><strong>Wild oat</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>diclofop-methyl</td>
<td>aryloxyphenoxypropionate</td>
<td>Post</td>
<td>563</td>
<td>Chemwet 1000 (0.25)</td>
</tr>
<tr>
<td>tralkoxydim</td>
<td>cyclohexanedione</td>
<td>Post</td>
<td>180</td>
<td>Supercharge (1.0)</td>
</tr>
<tr>
<td>mesosulfuron</td>
<td>sulfonylurea</td>
<td>Post</td>
<td>10</td>
<td>Chemwet 1000 (0.25)</td>
</tr>
<tr>
<td>triallate</td>
<td>thiocarbamate</td>
<td>Pre</td>
<td>800</td>
<td>-</td>
</tr>
<tr>
<td><strong>Both weeds</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>clethodim</td>
<td>cyclohexanedione</td>
<td>Post</td>
<td>60</td>
<td>Hasten (0.5)</td>
</tr>
<tr>
<td>glyphosate</td>
<td>glycine</td>
<td>Post</td>
<td>576</td>
<td>-</td>
</tr>
</tbody>
</table>
Table 2. Number of annual ryegrass populations collected that were herbicide resistant, developing resistance or susceptible from south-western New South Wales (% Total Resistant = resistant and developing resistant combined).

<table>
<thead>
<tr>
<th></th>
<th>Resistant</th>
<th>Developing resistance</th>
<th>Susceptible</th>
<th>Tested</th>
<th>% Total Resistant</th>
</tr>
</thead>
<tbody>
<tr>
<td>diclofop</td>
<td>53</td>
<td>16</td>
<td>55</td>
<td>124</td>
<td>56</td>
</tr>
<tr>
<td>sethoxydim*</td>
<td>38</td>
<td>10</td>
<td>21</td>
<td>69</td>
<td>70</td>
</tr>
<tr>
<td>clethodim</td>
<td>3</td>
<td>2</td>
<td>118</td>
<td>123</td>
<td>4</td>
</tr>
<tr>
<td>tralkoxydim</td>
<td>23</td>
<td>16</td>
<td>84</td>
<td>123</td>
<td>32</td>
</tr>
<tr>
<td>chlorsulfuron</td>
<td>49</td>
<td>14</td>
<td>57</td>
<td>120</td>
<td>53</td>
</tr>
<tr>
<td>imazamox/imazapyr</td>
<td>33</td>
<td>11</td>
<td>73</td>
<td>117</td>
<td>38</td>
</tr>
<tr>
<td>simazine</td>
<td>0</td>
<td>0</td>
<td>121</td>
<td>117</td>
<td>0</td>
</tr>
<tr>
<td>trifluralin</td>
<td>0</td>
<td>0</td>
<td>109</td>
<td>109</td>
<td>0</td>
</tr>
<tr>
<td>glyphosate</td>
<td>0</td>
<td>0</td>
<td>121</td>
<td>121</td>
<td>0</td>
</tr>
</tbody>
</table>

* sethoxydim was only screened to the diclofop resistant populations.

**Resistant:** diclofop, sethoxydim, clethodim, tralkoxydim, imazapic-imazapyr, glyphosate survival >20%; chlorsulfuron, simazine, trifluralin score >2.5).

**Developing resistance:** diclofop, sethoxydim, clethodim, tralkoxydim, imazapic-imazapyr, glyphosate survival 10-20%; chlorsulfuron, simazine, trifluralin score 1.5-2.5).

**Susceptible:** diclofop, sethoxydim, clethodim, tralkoxydim, imazapic-imazapyr, glyphosate survival <10%; chlorsulfuron, simazine, trifluralin score <1.5).
Table 3. Number of wild oat populations collected that were herbicide resistant, developing resistance or susceptible from south-western New South Wales (% Total Resistant = resistant and developing resistant combined).

<table>
<thead>
<tr>
<th></th>
<th>Resistant</th>
<th>Developing resistance</th>
<th>Susceptible</th>
<th>Tested</th>
<th>% Resistant</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>diclofop</td>
<td>20</td>
<td>18</td>
<td>66</td>
<td>104</td>
<td>37</td>
<td></td>
</tr>
<tr>
<td>clethodim</td>
<td>0</td>
<td>0</td>
<td>102</td>
<td>102</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>tralkoxydim</td>
<td>5</td>
<td>9</td>
<td>88</td>
<td>102</td>
<td>14</td>
<td></td>
</tr>
<tr>
<td>mesosulfuron</td>
<td>0</td>
<td>0</td>
<td>76</td>
<td>76</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>triallate</td>
<td>0</td>
<td>0</td>
<td>76</td>
<td>76</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>glyphosate</td>
<td>0</td>
<td>0</td>
<td>83</td>
<td>83</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>

Resistant: survival >20%, except for triallate score >2.5).
Developing resistance: survival 10-20%, except for triallate score 1.5-2.5).
Susceptible: survival <10%, except for triallate score <1.5).
Figure 1. Location of samples from four regions from the survey in south-western New South Wales showing resistance status of annual ryegrass populations to diclofop-methyl (a left) and chlorsulfuron (b right).