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Lessons learnt: crop-seed cleaning reduces weed-seed contamination in Western Australian grain samples

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Abstract. Weeds are a major contributing factor to crop yield loss. Weed control is regularly practiced during the growing season, with many growers making a conscious effort to minimise weed-seed return to the soil seedbank during the cropping program. However, growers may be unintentionally introducing weed seeds through sowing of contaminated crop seed. Using samples of crop seed obtained from 29 growers across two Western Australian grain-growing regions, 81 samples were hand-cleaned to determine weed-seed contamination levels. Of those samples, 41% were weed-free, and in the remaining 59%, the main contaminant was *Lolium rigidum* (annual ryegrass), occurring in 49% of contaminated samples. Crop type and cleaning method had significant effects on the level of weed-seed contamination, with barley having higher levels of contamination than other crops, and professional contractors providing lower contamination than other methods of cleaning. However, any seed-cleaning method provided significantly cleaner grain samples than no seed cleaning. This study established that crop-seed contamination was evident on Western Australian farms and that growers may be unintentionally sowing weed seeds with their crops. Seed cleaning combined with judicious paddock selection and weed-seed removal during the growing season can lead to weed-free crop seed.

Additional keywords: grain crops, surveys, herbicide resistance.

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Introduction

Weeds are a major factor contributing to yield loss in cropping systems (Jones *et al*. 2005), by interfering with crop development or by reducing the value of harvested crops. Weed seeds may contaminate harvested grain, with the potential for grain to be devalued or rejected. For example, weed seeds can taint wheat, leaving an undesirable odour that can affect the end quality of the produce (de Kantzow and Sutton 1988). Human-mediated seed dispersal has the potential to spread across large areas, with major sources of contamination being the movement of machinery during harvest and tillage operations (Blanco-Moreno *et al*. 2004; Barroso *et al*. 2006), livestock movement (Pleasant and Schlather 1994; Hogan and Phollips 2011), and sowing of contaminated grain (Pratley 2000; Moerkerk 2002; Michael *et al*. 2010). Many weed species are introduced through global commerce and grain traded between countries (Shimono and Konuma 2008; Shimono *et al*. 2010, 2015), although this risk of introducing weeds through grain is considered less than that from use of seed for planting (Wilson *et al*. 2016). However, the source or pathway of foreign seed movement changes for each stage (harvesting, grain cleaning, transport, storage and end use) from field to export, while the regulatory processes for these stages manage the risks associated with the introduction of foreign seed. Generally seed cleaning and import requirements reduce the risk of contamination (Wilson *et al*. 2016).

Herbicides are the preferred technology for weed control in many nations, and consequently, their continued and widespread use has seen the evolution of herbicide resistance (Heap 2019). In Australia, continuous cropping and no-till systems rely heavily on herbicides for weed control in broadacre cropping programs (Llewellyn *et al*. 2012). Resistance is now widespread in Australia for the major crop weeds, including *Lolium rigidum* Gaud. (annual ryegrass), *Raphanus raphanistrum* L. (wild radish), *Avena* spp. (wild oat) and *Bromus* spp. (brome grass) (Llewellyn and Powles 2001; Broster *et al*. 2011; Broster and Pratley 2006; Boutsalis *et al*. 2012; Owen *et al*. 2014, 2015*a*, 2015*b*; Owen and Powles 2016).

Weed-seed contamination in crop fields increases farm management costs, impacts on yield, and adds to cleaning costs of grain seed. It may also aid the spread of herbicide-resistant weeds and introduce unwanted foreign species into farming systems, with potentially serious consequences. For example, potential export markets may be threatened if weed-seed contaminants are affecting the quality of the end produce (such as flour) or have the potential to impact on local agricultural areas in the receiving country. Shimono *et al*. (2015) found that herbicide-resistant weed seeds had contaminated Australian grain samples exported to Japan, and that herbicide-resistant annual ryegrass plants had become established at most of the ports where the grain was delivered. Monitoring over a 3-year period revealed that the frequency of resistance was stable; however, resistant individuals were found up to 2 km from the ports, indicating that resistance was spreading over time. This herbicide-resistant weed seed is likely to spread into agricultural areas, eventually infesting local producers' crops.

Surveys in Western Australia have also shown that resistant weeds persist at harvest, although weed densities are generally low (<1 plant m^{-2}) (Llewellyn *et al*. 2009; Owen *et al*. 2014). However, each plant can produce large quantities of seeds, which are likely to end up as contaminants of harvested grain. Previous studies have identified weed-seed contaminants in harvested grain and crop seed samples (Powles and Cawthray 1999; Niknam *et al*. 2002; Shimono and Konuma 2008; Michael *et al*. 2010; Shimono *et al*. 2015). In Australia, growers regularly employ in-crop weed management practices to reduce weed seedset, thus minimising the number of seeds returning to the seedbank, and many clean their harvested grain by using a variety of seed-cleaning operations (Michael *et al*. 2010). If seed-cleaning operations are only partially effective, then weed seeds may be sown into fields along with the crop seed in the following season or exported with grain delivered to market. Minimising the introduction of weeds into the farming system through sowing of clean crop seed is an important component of best farming practice.

In 2008, Michael *et al*. (2010) studied the extent of weed-seed contamination in retained crop seed used for sowing by Western Australian grain growers. This subsequent study compares the current levels of weed-seed contamination in crop grain with those of the previous study and explores whether farmers are changing their focus on seed-cleaning techniques.

Materials and methods

Grain collection

During March–May 2015, 81 grain samples were collected from 29 growers across two regions of the Western Australian grain belt (Fig. 1). These growers participated in the aforementioned earlier survey (Michael *et al*. 2010). Each

Fig. 1. Map of the Western Australian grain belt, indicating the two regions (Dalwallinu and Corrigin) and 29 locations where grain samples were collected between March and May 2015. In total, 81 samples were collected from the two regions, 35 at Dalwallinu, 46 at Corrigin.

grower provided cleaned crop-seed samples (~10 kg) of commonly used varieties of wheat (*Triticum aestivum* L.), barley (*Hordeum vulgare* L.), rapeseed (*Brassica napus* L.), and/or lupin (*Lupinus* spp.). In addition, eight uncleaned crop samples (also collected from growers) were used as controls to demonstrate the effects of seed cleaning on weed-seed contamination. Crop management practices such as cleaning methods of crop seed, source of crop seed (grain source), crop type and variety, and changes to seed-cleaning methods since the 2008 survey were investigated. Growers were also asked to estimate the expected level of weed-seed contamination in the grain samples provided. Cleaning methods of crop seed were categorised into groups based on method and location: 'in town', by external contractors using gravity tables; 'mobile', by external contractors using rotary screens and/or sieves at the farm; and 'home', by farmers using sieves and air. This contrasts with our previous study (Michael *et al*. 2010) in which grain-cleaning methods were categorised by implement used (gravity table, rotary screen, sieve, and combination). The present classification allows a fairer division between cleaning methods, because all of the 'town' cleaners used gravity tables, whereas there was some variation between the types of cleaning within the mobile units. There was also uncertainty among growers when defining the cleaning implement used in some mobile cleaning units. On average, each sample of grain collected weighed 11 kg. All grain samples (81 cleaned and eight uncleaned) were then further hand-cleaned by using a combination of sieves (2.8, 2.0 and 1.0 mm) and hand-picking in order to remove all foreign contaminants and determine the quantity of weed-seed contamination. Weed seeds were quantified and separated by species. Seeds of the four major weeds (annual ryegrass, wild radish, wild oat and brome grass) found in the grain samples were then germinated and the seedlings screened for resistance to the most commonly used selective herbicides (e.g. diclofop, chlorsulfuron, fenoxaprop) for weed control (Table 1). In order to calculate a reliable percentage survival, only those samples consisting of >10 weed seeds were screened. At least two species of each of the genera *Avena*, *Bromus* and *Hordeum* (barley grass) are known to co-exist in the Western Australian grain belt, with many fields containing both species (Owen *et al*. 2015*a*, Owen and Powles 2016). The common species are *A. fatua* L. and *A. sterilis* subsp. *ludoviciana* Durieu; *B. diandrus* Roth and *B. rigidus* Roth; and *H. glaucum* Steud. and *H. leporinum* Link. For the purposes of this study, all species within a genus were considered a single type of weed; hence, 'wild oat' refers to *Avena* spp., 'brome grass' to *Bromus* spp., and 'barley grass' to *Hordeum* spp.

Herbicide-resistance testing of contaminating crop seed

During August and September 2015, the recovered weed seeds from the crop grain samples (both cleaned and uncleaned) were placed in 500-mL plastic containers containing 1% (w/v) solidified agar-water. Ryegrass seeds were germinated in a growth cabinet with alternating 12-h periods of light at 25°C and dark at 15°C for 5 days, while wild oat and brome grass seeds were placed in the fridge at 5°C for 7 days. Seedlings (10 per sample) were then transplanted into plastic trays (30 cm by 40 cm by 10 cm) containing potting mix (50% composted pine bark, 25% peat and 25% river sand). Wild Radish seeds were sown directly into the seed trays containing potting mix. Trays were kept outdoors at the University of Western Australia, Perth, and watered and fertilised as required. Seedlings were treated at the two- to three-leaf stage with herbicide at the upper recommended field rates (Table 1) together with wetting agent as required, using a custom-built, dual-nozzle (TeeJet XR11001 flat fan; TeeJet Technologies, Wheaton, IL, USA) cabinet sprayer delivering herbicide in water at a rate of 110 L ha^{-1} at 200 kPa, and a speed of 3.6 km h^{-1}. Herbicides commonly used to control each weed species were used. Known herbicide-resistant and herbicide-susceptible populations were used as controls for each herbicide treatment (Owen *et al*. 2014, 2015*a*, 2015*b*; Owen and Powles 2016). In all experiments, 100% mortality occurred in the known susceptible populations, whereas there was always very high survival (>90%) in the known resistant populations for all herbicides used. The effect of the herbicide was assessed by determining seedling mortality 21 days after herbicide treatment. Weed samples were classed as resistant if any plants survived treatment, and susceptible if all plants died.

Data analyses

All foreign seed contaminants (annual ryegrass, wild radish, brome grass, wild oat and volunteer crop species) were analysed. A linear mixed model was used to examine the effects of cleaning method (mobile, in town, home) and location (Corrigin, Dalwallinu; Fig. 1), grain source (i.e. home grown or from external sources), crop type, crop variety, and region (as fixed effects) on the square-root of total average number of seeds, with 'farmer' included as a random effect in the model. Non-significant factors were

Table 1. Herbicide-resistance status of weed seeds contaminating crop seed samples, showing the average number of seeds tested and average plant survival per sample

Weed species	Herbicide	Herbicide rate (g a.i. ha^{-1})	No. of samples tested	Av. no. of seeds tested	Av. survival (%)	No. of samples with resistance
Annual ryegrass	Diclofop	500	19	35	75	19
	Sulfometuron	15	8	42	83	8
Wild radish	Chlorsulfuron	10	3	10	96	3
Wild oat	Fenoxaprop	38.5	2	15	0	0
Brome grass	Fluazifop	78	1	50	0	0

removed, and the effects of the remaining factors were estimated from the model. A square-root transformation was required to ensure that statistical assumptions in the model were valid. GENSTAT Edition 18 (VSN International, Hemel Hempstead, UK) was used for all data analyses.

Results

Of the 81 cleaned crop samples, 37 were wheat, 23 barley, 8 canola and 13 lupin. The uncleaned samples comprised a mix of five cereal (wheat (1), barley (2) or oat (2)) and three canola crops. Nearly all grain samples (97%) were sourced from within the farm (home-grown), with the remainder purchased from seed agents or local growers. Despite most growers retaining their own crop seed (home-grown), it was primarily cleaned by external grain cleaners rather than by the farmer. Three grain-cleaning categories (see *Methods*) were used by the farmers surveyed, with use of local seed cleaners 'in town' being most common (81%), followed by 'mobile' seed cleaners on-farm (14%), with 5% of farmers cleaning their seeds 'at home'.

Crop-seed contamination

Of all cleaned crop seed samples, 59% had some level of weed-seed contamination, with an average of 18 ± 7 weed seeds in a 10-kg grain sample across all crops (Table 2). This was close to farmer predictions of 21 ± 3 weeds seeds per 10-kg grain sample.

Seeds of 13 different species contaminated the crop seed (Fig. 2, Table 2), comprising 11 agricultural weed species and two volunteer crop species. The most common weed contaminant was annual ryegrass, which occurred in almost half of the collected samples (49%), followed by wild radish (16%), wild oats (12%) and brome (11%). These species also had the highest levels of contamination, at 14 seeds (annual ryegrass), 2 seeds (wild radish), 1 seed (wild oats) and <1 seed (brome) per 10-kg sample. The remaining weed species were found in 7% of samples with an average of <1 seed per 10-kg grain sample. Many samples (74%) were contaminated with volunteer cereal (54%) and/or legume (43%) grains. Volunteer crop species, mainly wheat in barley samples, accounted for high levels of contamination, averaging 16 ± 8 seeds per 10-kg crop-seed sample (Fig. 2). Two samples contained unidentified weed species (accounting for <1% total contamination) (Table 2); however, these seeds could not be germinated and consequently the species were unable to be identified.

There were clear differences in the level of weed-seed contamination between crops, with contamination in barley crops being significantly ($P < 0.05$) higher than in other crops. The level of weed-seed contamination was variable (no. of seeds per 10-kg crop-seed sample: barley 47 ± 22, canola 14 ± 5, wheat 7 ± 3, lupin, <1 ± 1) (Table 3). The non-leguminous crops had a higher incidence of weed-seed

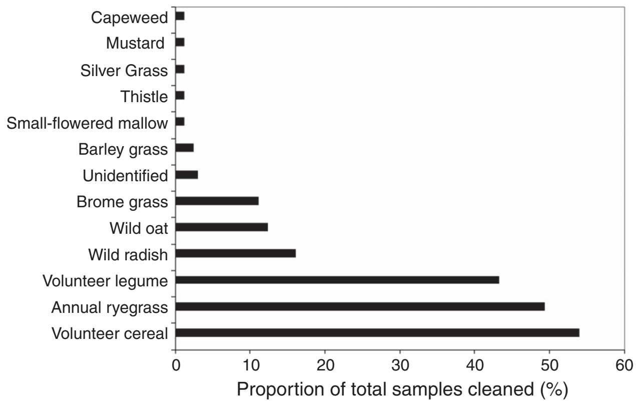

Fig. 2. Common seed contaminants as a proportion of the total cleaned samples. Eleven species of weed-seed contaminants were identified, with the remaining contaminants being volunteer crop species.

Table 2. Contamination levels (samples contaminated, SC) and average number of weed seeds (± standard error) in a 10-kg sample of cleaned grain
In total, 81 samples were collected

Common name	Scientific name	All crops		Wheat		Barley		Lupin		Canola	
		% SC	No. of seeds	% SC	No. of seeds	% SC	No. of seeds	% SC	No. of seeds	% SC	No. of seeds
Annual ryegrass	*Lolium rigidum* Gaud.	49	14 ± 6	54	5 ± 2	65	37 ± 21	0	0 ± 0	63	12 ± 5
Wild radish	*Raphanus raphanistrum* L.	16	2 ± 1	8	<1.0 ± 0.2	26	5 ± 3	15	<1.0 ± 0.3	25	<1.0 ± 0.7
Brome grass	*Bromus* spp.	11	<1.0 ± 0.2	14	<1.0 ± 0.1	17	1 ± 0.5	0	0 ± 0	0	0 ± 0
Wild oat	*Avena* spp.	12	1 ± 0.8	11	<1.0 ± 0.1	22	3 ± 3	8	<1.0 ± 0.1	0	0 ± 0
Barley grass	*Hordeum* spp.	2	<1.0 ± 0.1	3	<1.0 ± 0.1	4	<1.0 ± 0.1	0	0 ± 0	0	0 ± 0
Small-flowered mallow	*Malva parviflora* L.	1	<1.0 ± 0.1	0	0 ± 0	4	0 ± 0	0	0 ± 0	0	0 ± 0
Thistle flat weed	*Hypochaeris radicata* L.	1	<1.0 ± 0.2	3	<1.0 ± 0.1	0	0 ± 0	0	0 ± 0	0	0 ± 0
Silver grass	*Vulpia* spp.	1	<1.0 ± 0.2	0	0 ± 0	4	<1.0 ± 0.1	0	0 ± 0	0	0 ± 0
Wild mustard	*Sisymbrium officinale* L.	1	<1.0 ± 0.5	3	<1.0 ± 0.5	0	0 ± 0	0	0 ± 0	0	0 ± 0
Capeweed	*Arctotheca calendula* (L.) K.Lewin	1	<1.0 ± 0.2	0	0 ± 0	0	0 ± 0	8	<1.0 ± 0.1	0	0 ± 0
Unidentified		1	<1.0 ± 0.2	3	<1.0 ± 0.1	0	0 ± 0	0	0 ± 0	12	<1.0 ± 0.6
Volunteer cereal		54	14 ± 8	49	2 ± 1	43	40 ± 26	92	4 ± 0.1	50	3 ± 2
Volunteer legume		43	2 ± 0.6	46	2 ± 0.7	48	3 ± 1	31	3 ± 2	38	2 ± 0.8
Total weeds		59	18 ± 7	62	7 ± 3	74	47 ± 22	15	1 ± 1	75	14 ± 5
Total volunteer crops		74	16 ± 8	73	4 ± 2	65	43 ± 27	92	7 ± 2	75	5 ± 1
Total contamination		88	34 ± 10	84	11 ± 4	87	90 ± 4	92	8 ± 2	100	19 ± 5

Table 3. Changes in weed-seed incidence and contamination levels in cleaned grain samples between 2008 and 2015 (± standard errors)
Data for 2008 from Michael *et al*. (2010). $P < 0.05$ considered significant

Factor	2008	2015	*P*-value
Samples cleaned by independent contractor (%)	71	95	
Samples cleaned by farmer (%)	29	5	
Samples contaminated with weed seed (%)	73	59	
Samples contaminated with annual ryegrass (%)	45	49	
Samples contaminated with wild radish (%)	30	16	
Samples contaminated with brome grass (%)	29	11	
Samples contaminated with wild oat (%)	24	12	
Samples contaminated by volunteers (%)	27	74	
Samples with contamination (weed and volunteer) (%)	73	88	
Weed-seed contamination level (seeds per 10-kg sample)	56 ± 11	18 ± 7	0.001
Total contamination (weed and volunteer crop seeds per 10-kg sample)	62 ± 11	34 ± 10	0.018
Contamination in samples cleaned by independent contractors (weed seeds per 10-kg sample)	50 ± 12	11 ± 4	0.003
Contamination in samples cleaned by farmers (weed seeds per 10-kg sample)	91 ± 24	144 ± 100	0.405
Weed seed contamination in wheat (seeds per 10-kg sample)	61 ± 15	7 ± 3	0.004
Weed seed contamination in barley (seeds per 10-kg sample)	53 ± 21	47 ± 22	0.793
Weed seed contamination in lupin (seeds per 10-kg sample)	43 ± 16	1 ± 1	0.129
Weed seed contamination in canola (seeds per 10-kg sample)	57 ± 20	14 ± 5	0.083
Annual ryegrass contamination (seeds per 10-kg sample)	28 ± 5	14 ± 6	0.099
Wild radish contamination (seeds per 10-kg sample)	14 ± 5	2 ± 1	0.007
Brome grass contamination (seeds per 10-kg sample)	9 ± 4	<1.0 ± 0.2	0.002
Wild oat contamination (seeds per 10-kg sample)	4 ± 2	1 ± 0.8	0.034

contamination than lupin (15% of samples): barley 74%, wheat 62%, canola 75%. In addition, some barley crop varieties had higher levels of weed-seed contamination than others. There were seven barley varieties, which included Hindmarsh, Buloke, Litmus, Scope, Latrobe, Fathom, and Yagan. The variety Buloke was significantly ($P < 0.001$) more contaminated than all other varieties at 44 seeds per 10-kg grain sample, and Scope also had high levels of weed-seed contamination (8 seeds per 10-kg grain sample); all other varieties had <2 seeds per 10-kg grain sample. Crop variety did not significantly affect levels of weed-seed contamination in the other crop species, even though there were also seven varieties of wheat and six of lupin.

The most common weed-seed contaminant in wheat was annual ryegrass (in 54% of samples), followed by brome (14%), wild oat (11%) and wild radish (8%). Annual ryegrass is also the most commonly found weed in grain-growing regions in southern Australia. It also had the highest contamination levels of 5 seeds per 10-kg grain sample. The other grass weed species (brome, barley grass, wild oat) had <1 seed per 10-kg grain sample (Table 2). Annual ryegrass was also the most common contaminant in barley (65%) and canola (63%), while none was found in lupin samples. Whereas brome grass was found in wheat (<1 seed per 10-kg sample) and in barley (17%, 1 seed per 10-kg sample), it was not found in canola and lupin crops. Wild oat, as well as being present in wheat (<1 seed per 10-kg sample), was also present in barley (22%, 3 seeds per 10-kg sample) and lupin (8%, <1 seed per 10-kg sample). Wild radish was present in all crops: wheat <1 seed per 10-kg sample; barley (26%), 5 seeds per 10-kg sample; canola (25%), 1 seed per 10-kg sample; lupin (15%), <1 seed 10-kg sample (Table 2).

Table 4. Contamination levels and average number of weed seeds (± standard error) in a 10-kg sample of uncleaned grain
Eight samples were collected

Common name	Contamination (% of samples)	No. of seeds
Annual ryegrass	100	1790 ± 1056
Wild radish	50	25 ± 18
Brome grass	50	22 ± 22
Wild oat	50	4 ± 1
Barley grass	13	2 ± 1
Volunteer cereal	75	88 ± 41
Volunteer legume	25	7 ± 6
Total weeds	100	1841 ± 1050
Total volunteer crops	88	95 ± 45
Total contamination	100	1936 ± 1046

Crop cleaning and management practices

Method of seed cleaning had a significant ($P < 0.001$) effect on the level of weed-seed contamination. Farmer-cleaned samples had significantly higher levels of weed-seed contamination (144 ± 100 seeds per 10-kg sample) than samples cleaned by independent contractors, either mobile (using a combination of sieves and screens) or in-town (using gravity tables). Samples cleaned in town contained 10 ± 4 weed seeds per 10-kg grain sample, which was not significantly different from samples cleaned by mobile seed cleaners (17 ± 11), although there was a trend for in-town cleaners to have less weed-seed contamination. Any form of seed cleaning was beneficial, with the uncleaned control samples containing 1841 ± 1050 weed seeds in a 10-kg sample (Table 4) (32 times higher than the overall average of the cleaned samples; $P < 0.001$).

The efficacy of the cleaning method was not affected by weed species, except in the case of wild radish. In-town cleaners, with 1.0 ± 0.5 weed seed per 10-kg crop sample, had significantly ($P < 0.05$) lower wild radish contamination levels than mobile cleaners (3.0 ± 1.5 weed seeds per 10-kg crop sample), which in turn had significantly lower levels than cleaning methods employed on farm (8 ± 2 weed seeds per 10-kg crop sample). Barley crops tended to have higher wild radish contamination than wheat and lupin crops. Barley crops also had greater levels of contamination in general.

Region from which the samples were collected had no significant effect on the level of weed-seed contamination. Although, generally, the grain source also was not significant, brome grass (2.0 ± 0.5 weed seeds per 10-kg crop sample) and wild oat (15 ± 3) contamination levels were significantly higher ($P < 0.05$ and $P < 0.001$, respectively) in grain samples that had been brought onto the farm (the origin of these samples was not investigated) than those sourced from within the farm (<1 ± 1 seed per 10-kg crop sample). Farmer-cleaned grain samples contained significantly more brome and wild oat seeds than samples from commercial cleaners.

Herbicide resistance

Weed seeds found contaminating the crop-seed samples (both cleaned and uncleaned) were germinated and screened for resistance to commonly used herbicides (Table 1). Most annual ryegrass individuals found in the crop seed were resistant to the selective in-crop herbicides diclofop-methyl and sulfometuron, and wild radish had resistance to chlorsulfuron. By contrast, no resistance to fenoxaprop or fluazifop was found in wild oats or brome grass, respectively.

Changes since 2008

The proportion of weed-free crop-seed samples increased from 27% in 2008 to 41% in 2015, indicating that growers are thinking about the cleanliness of their retained crop seed. Use of external seed cleaners, in particular the in-town seed cleaners, increased from 71% of farmers in 2008 to 95% in 2015. Overall, the level of weed-seed contamination decreased ($P < 0.001$) from 56 ± 11 seeds per 10-kg grain sample in 2008 to 18 ± 7 in 2015, with annual ryegrass remaining the main weed contaminant. However, the incidence of volunteer crops (particularly wheat in barley samples) increased from 27% to 74% of contaminated samples. Overall contamination (weed and volunteer crop seed) also reduced from 62 ± 11 to 34 ± 10 seeds per 10-kg grain sample ($P < 0.05$) during this period (Table 3). Grain samples cleaned by independent contractors had 11 ± 4 seeds per 10-kg sample in 2015, which was significantly less ($P < 0.01$) contamination than in 2008 (50 ± 12 seeds per 10-kg sample; Table 3). Weed-seed contamination levels in barley crops were similar for both surveys; however, contamination was significantly lower for wheat crops in 2015 (Table 3). The incidence of sample contamination with annual ryegrass decreased from 28 ± 5 seeds in 2008 to 14 ± 6 seeds in 2015, and there was also a significant decrease in contamination levels of other weed species such as brome grass, wild radish and wild oat in 2015 (Table 3).

Discussion

Our study confirms that although farmers are still introducing weeds and volunteer crops into their farming systems, seed-cleaning techniques are used extensively and there was a 14% increase in the number of weed-seed-free grain samples compared with our previous study in 2008 (Michael *et al*. 2010). Similarly, the level of weed-seed contamination per sample has decreased since 2008 for all weed species. In 2008, the level of weed-seed contamination was 56 seeds per 10-kg grain sample, equating to 465 weed seeds ha^{-1} (Michael *et al*. 2010). In 2015, the level of weed seed contamination was 18 seeds per 10-kg sample (135 weed seeds ha^{-1}), and thus the chance of introducing weed-seed contaminants in the following year would be much lower. This is encouraging, because as was the case in 2008, many of the major contaminating weed seeds were resistant to commonly used post-emergent herbicides. Recent random surveys have shown that herbicide resistance levels have increased in cropping fields, particularly in annual ryegrass and wild radish resistant to post-emergent in-crop herbicides (Owen *et al*. 2014, 2015*a*, 2015*b*; Owen and Powles 2016); therefore, returning herbicide-resistant seeds to the weed seedbank during crop sowing would exacerbate the problem.

The most common weed-seed contaminants in crop grain in 2015 were the same as those in 2008 (Michael *et al*. 2010), namely annual ryegrass, wild radish, wild oat and brome grass, with ryegrass infesting a similar proportion of crop samples in both years, and the incidence of the other species having decreased by ~50% or more (Table 3). Borger *et al*. (2012) found that dominance of weed species in Western Australia had changed over the period between 1997 and 2008, and thus changing farming practices could be favouring certain weed species over others. In particular, *Bromus diandrus* showed a significant decrease in incidence in cropped fields, and this was reflected in lower levels of *Bromus* seed contamination in the 2015 grain samples.

Crop type had a major effect on the level of weed-seed contamination in both studies. In the 2008 study, barley crops were significantly cleaner than other crop types (Michael *et al*. 2010) but the opposite was true in 2015. Although contamination levels in barley crops were similar in both surveys, wheat and lupins crops had considerably less contamination in 2015. This could be due to the variety of barley used and/or the seed-cleaning method employed. In 2015, varieties Buloke and Scope had higher levels of weed-seed contamination than the other barley varieties and accounted for 30% of barley samples (note that barley samples were not separated into varieties in the 2008 study). Anecdotally, Buloke and Scope are harder to clean than other varieties because of their size, weight and shape (Bruce Rock Seed Cleaners 2016, pers. comm.). Additionally, most of the grain samples cleaned by the farmer at home (the least efficient method of cleaning) were barley, which may also partly explain the higher levels of weed-seed contamination seen in barley crops. Interestingly, more samples were contaminated with volunteer cereals in 2015 than in 2008, although weed-seed numbers were much lower, which could be due to the previous season's crop germinating in the following year.

Although most growers stated that they had not substantially changed their cleaning methods since 2008, many had changed the way they sourced their crop seed, i.e. by selecting cleaner paddocks with a low weed-seed burden and practicing good on-farm hygiene such as thorough cleaning of sheds and silos to reduce weed-seed contamination. The use of external contractors had also increased from 81% to 95%. These changes had a positive effect on the average number of weeds seeds per 10-kg crop sample, dropping from 56 to 18 seeds over the 7 years between studies. Overall, more growers were using in-town cleaners in 2015 rather than mobile contractors or self-cleaning. Although there was no significant difference in the overall level of weed-seed contamination in grain samples cleaned by external contractors in town and in mobile units, some differences in seed-contamination levels between individual mobile seed cleaning units can be accounted for by the operator or user; for example, faster operating speeds usually result in less weed-seed removal. The type of unit fitted to the mobile cleaner can also affect efficiency, with new cleaning units having more efficient systems (Corrigin Seed Cleaners 2016, pers. comm.).

Most growers retained their own seed rather than buying certified seed from external sources. Although certified seed guarantees purity in terms of variety and quality, it does not guarantee the seed to be weed-free. Regulations covering the sale of crop seed may not totally exclude foreign seeds and there are no requirements to declare the herbicide-resistance status of any contaminating seeds (Matthews 1994). There are acceptable limits for weed-seed contaminants based on foreign-material assessment for the delivery of grain to local bulk handling facilities (i.e. Co-operative Bulk Handling, CBH). Weed seeds are categorised by type and have individual limits for each grade. For example, <50 seeds of common weeds such as annual ryegrass and wild radish and volunteer cereals are allowable per sample. Noxious weeds and tainting agents have a non-tolerable limit and a penalty of non-acceptance, whereas other weeds have a tolerable limit which changes depending on the species and may also attract a price-discount penalty (de Kantzow and Sutton 1988; CBH 2019).

As discussed in our earlier work (Michael *et al*. 2010), weed-seed contamination can pose a risk to both local and export markets, particularly in major wheat-importing countries such as Japan, which consequently means that large numbers of weed seeds are introduced to these international trading ports. Shimono *et al*. (2015) found that grain delivered to ports in Japan from Australia, Canada and the United States contained herbicide-resistant weed seed. At least some of this seed is then likely to be spread and naturalise into agricultural areas throughout Japan. This highlights the importance of considering where crop grain is sourced, because it has the potential to bring unwanted, invasive plants into agricultural systems with potentially devastating effects. International grain-trade pathways represent a significant contribution to new pests and diseases around the world (Wilson *et al*. 2016). Regulating this spread is the responsibility of individual countries under International Plant Protection Convention (IPPC) guidelines involving legislation and import requirements to reduce the risk. In general, crop–weed associations at the point of grain origin, farming practices, grain handling, transport and storage, import requirements, and the end use of the grain all need to be considered when assessing the pathway risk of introducing foreign seeds (Wilson *et al*. 2016).

Utilising thorough seed-cleaning and crop-hygiene practices will assist in reducing weed-seed contamination levels in crop seed and thereby decrease the risk of sowing herbicide-resistant weed seeds back into cropping fields. The level of weed-seed contamination can be reduced if farmers choose fields with a low weed burden for crop seed production, as well as adhering to good on-farm hygiene practices (clean silos, trucks and equipment, monitoring of livestock movement), because contaminated equipment, particularly at harvest and grain handling, is the usual source of introducing weed seeds (Matthews 1994). Delaying crop sowing to allow adequate weed control could also reduce the end-of-year weed burden for future crop plantings. Harvest weed-seed control systems that target weed seeds in the chaff fraction during crop harvest, and thus prevent seed from re-infesting the field, help to reduce the weed burden in the following years (Walsh *et al*. 2013).

Crop type is also likely to influence contamination levels. Some crops and varieties are more competitive and allow a greater choice of herbicides for in-crop weed control, whereas some are more difficult to clean at the end stage. It is important to monitor weed-seed contamination levels at sowing and during harvest to ensure that the cleanest possible crop seed is available while not affecting in-crop weed control during the year. The impact of crop-cleaning systems on crop-seed contamination and the potential introduction of herbicide-resistant weed populations, particularly in areas where there are limited options for weed control during the growing season, need to be considered. In the end, all techniques that prevent weed seeds from returning to the soil seedbank, whether chemical, management-based or mechanical, are required to reduce the yearly weed burden and maintain the sustainability of farming systems in the future.

Conflicts of interest

The authors declare no conflicts of interest.

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