

## Ecology of threehorn bedstraw (*Galium tricornerutum*): Implications for management and harvest weed seed control

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**Summary** Threehorn bedstraw is an important broadleaf weed of crops in southern Australia but its behaviour in field crops has not been adequately investigated. Populations of bedstraw ( $n = 10$ ) were collected from the southern cropping region to investigate their seed dormancy and seedling emergence pattern. Seeds of these populations were placed outdoors in seedling trays in autumn and watered as needed until spring to prevent water stress. There was no variation in the pattern of seedling emergence in these populations but their cumulative germination ranged from 14–46% of the initial seedbank. Presence of such high levels of ungerminated seedbank in bedstraw is consistent with previous reports of its high seedbank persistence. Seedling emergence in these populations only occurred after the onset of cold conditions in late winter (July–August), which is consistent with stimulation of seed germination by cold-stratification. Competitive effects of varying bedstraw densities were investigated in wheat (cv. CLF-Hatchet and Sceptre) and lentils (cv. Hurricane XTA). There was a hyperbolic relationship between bedstraw density and its seed production ( $R^2 = 0.82–0.93$ ) in both crops, however seed production was much higher in lentils ( $>20,000$  seeds  $m^{-2}$ ) than wheat (3800–5900 seeds  $m^{-2}$ ). Despite its late seedling emergence, there was an exponential relationship between bedstraw density and yield loss in wheat and lentil. Maximum yield loss relative to the weed-free control was 10–14% in wheat and  $\geq 30\%$  in lentils. These results highlight the need to develop effective bedstraw management tactics, especially in lentils. Bedstraw showed no seed dispersal prior to the harvest-ready stage of wheat and lentil (100% retained). Therefore, harvest weed seed control practices including seed catchers, weed seed destructor and narrow windrow burning could be highly effective in its long-term management provided seeds could be separated into the chaff fraction in the harvester.

**Keywords** Ecology.

### INTRODUCTION

Threehorn bedstraw (*Galium tricornerutum* Dandy) is an important broadleaf weed of winter crops in southern Australia. This weed has become more prevalent

across the Wimmera district of Victoria, and mid north and Yorke Peninsula of South Australia where lentil area has increased considerably over the last 10 years. Lentils are weak competitors with weeds (McDonald *et al.* 2007), and at present there are few herbicide options for the selective control of this weed in lentils.

Despite increasing concerns about threehorn bedstraw in southern Australian farming systems, there are significant knowledge gaps in its seed biology. Chauhan *et al.* (2006a, b) reported that germination of threehorn bedstraw was inhibited by light, but stimulated by exposure to cold-stratification or treatment with gibberellic acid. According to Mennan (2003), seed persistence in catchweed bedstraw (*Galium aparine*) was limited to 2–3 years in the field but could be increased by deep burial. Catchweed bedstraw is a competitive weed of field crops, which can reduce cereal yields by 30–60% (Rola 1971). Seeds of catchweed bedstraw were found to be a significant contaminant of cereal grain, and it was shown to increase crop lodging by smothering crops, which made harvest more difficult (Defelice 2002).

Even though threehorn bedstraw is an important weed in southern Australia, there is little information available on its seedbank persistence and crop-weed interference. At present there is considerable interest in the grains industry in harvest weed seed control (HWSC) tactics including weed seed catchers, weed seed destructor and narrow windrow burning. The effectiveness of these practices depends largely on the timing of weed seed dispersal and the amount of weed seed present above the cutter bar height at crop harvest (Walsh and Powles 2014). However, there is no information available on seed production, retention and shedding behaviour of threehorn bedstraw, which will strongly influence prospects of HWSC for its management.

More information on seedling emergence pattern, seed persistence, seed production, retention, and shedding behaviour of threehorn bedstraw is needed to facilitate development of effective control strategies. Studies were therefore undertaken to address these knowledge gaps.

## MATERIALS AND METHODS

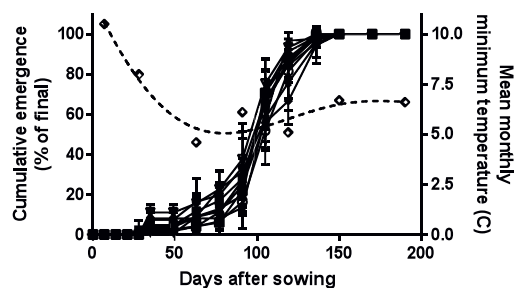
A field study was undertaken in 2017 at Roseworthy (34°32' S, 138°41' E), South Australia, to investigate field seedling emergence and seedbank recruitment of 10 populations of bedstraw randomly collected from cropping fields in Victoria and South Australia. Seeds were collected before crop harvest in October to November 2016 from more than 200 mature plants at each site, and were immediately stored outdoors in naturally lit storage units to maintain ambient temperature conditions until used for experiments. In the autumn after seed collection (March 2017), 100 seeds of each population ( $n = 10$ ) were sown in plastic trays (33 cm by 28 cm by 5 cm) containing potting mix and were maintained outside during the normal growing season at Roseworthy. There were four replicate trays of each population. Trays were watered as required to maintain good soil moisture conditions for seed germination and seedling emergence. Weed seedlings were counted (at first leaf appearance) from April to the end of October (7 months or 190 d). Initially, emergence was recorded at weekly intervals but after 1.5 months, because of low emergence, it was recorded at 2-week intervals. Seedlings were counted and removed until no further emergence was recorded in three consecutive measurements. The counts were expressed as cumulative seedling emergence (i.e. percentage of the total emergence). Cumulative seedling emergence data were fitted using a sigmoidal logistics model (GraphPad Prism v.6.0; GraphPad Software, San Diego, California) to compare the time taken (d) by populations to reach 50% maximum seedling emergence ( $t_{50}$ ).

Additional field studies were undertaken at Roseworthy to determine seed production, crop yield loss and pattern of seed dispersal of bedstraw in wheat (2016 and 2017) and lentils (2017). Plots of nil, low (10–20 plants  $m^{-2}$ ), medium (50 plants  $m^{-2}$ ), and high ( $\geq 100$  plants  $m^{-2}$ ) weed density of bedstraw were established by hand-broadcasting weed seeds prior to sowing wheat and lentils. In 2016, the experiment was planted with wheat cv. CLF-Hatchet at 100 kg  $ha^{-1}$  seed rate on 6 June, whereas wheat cv. Sceptre at 100 kg  $ha^{-1}$  and lentil cv. Hurricane XTA at 45 kg  $ha^{-1}$  seed rates were planted on 7 and 8 June in 2017. Diammonium phosphate required to supply 22.5 kg N  $ha^{-1}$  and 25 kg P  $ha^{-1}$  was banded below the crop seed at sowing. Plots (5 m long and 1.5 m wide) were sown using a knife-point and press wheel seed drill on 0.25 m row spacing, and both crops were managed according to the district practice. The experiments were established as a randomised complete block design with four replicates. Weed seed production of bedstraw was assessed within four weeks of crop senescence

in November 2016 and 2017. Plant density and seed production was assessed in two quadrats (0.5 m by 0.5 m) in each plot. A hyperbolic model was found to best fit the relationship between weed density and weed seed production. High density plots of bedstraw were also used to measure temporal pattern of weed seed dispersal. At seven weeks before the expected harvest-ready stage of the crop ( $\leq 12\%$  grain moisture content), three circular seed traps (15 cm diam. by 10 cm deep) were randomly placed between crop rows in each plot. A wire screen was fitted to the bottom of each trap to capture seed rain. Eight holes at the bottom of the seed trap allowed rainwater drainage. Weed seeds were collected weekly from the traps until just before crop harvest. Collected seeds were identified and counted to quantify seed dispersal. Grain yield was determined using a Kew plot harvester, and a one-phase exponential decay model was fitted to the data to assess the impact of weed density on grain yield. Contamination of grain sample by weed seeds at harvest was determined by counting the number of bedstraw seeds in a grain subsample ( $\sim 500$  g) from each plot.

## RESULTS

All populations exhibited no seedling emergence until 28 DAS (Figure 1). Furthermore, the time taken to reach 50% of maximum seedling emergence was similar across the populations and ranged from 95–106 days (Table 1). The start of seedling emergence coincided with minimum air temperatures dropping below 5°C in the field (Figure 1). Cumulative seedling emergence from the seedbank did not exceed 50% for any of the populations, and ranged from 14–46% (Table 1). These results indicate large persistence of bedstraw seedbank from one year to the next.



**Figure 1.** Cumulative seedling emergence pattern of bedstraw populations ( $n = 10$ ) collected from Victoria and South Australia. Dashed line represents mean monthly minimum temperature, and vertical bars are the SE of the mean.

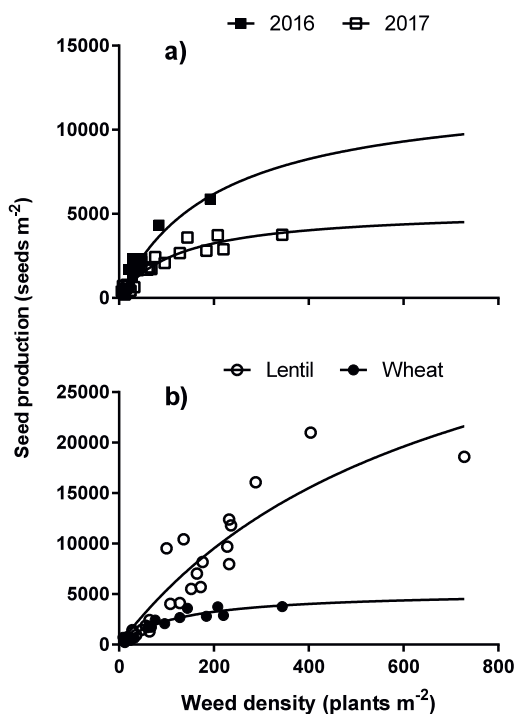
**Table 1.** Cumulative seedling emergence from the seedbank and time taken (days) to reach 50% of maximum seedling emergence ( $t_{50}$ ) of bedstraw populations from Victoria and South Australia. Values in parenthesis represent SE of mean, and  $t_{50}$  values were calculated by fitting cumulative emergence data to a sigmoidal logistic model.

Population	Cumulative seedling emergence (% of seedbank)	Time taken (d) to reach 50% seedling emergence ( $t_{50}$ )	$r^2$
BE2 YP	32 (3.6)	98.1	0.95
BE5 SE	38 (4.0)	99.9	0.99
BE9 WD	14 (3.8)	99.2	0.97
BE14 YP	45 (2.6)	99.5	0.99
BE15 YP	31 (2.6)	105.6	0.98
BE16 YP	36 (4.1)	105.1	0.99
BE17 MN	46 (3.7)	99.6	0.99
BE18 SE	27 (3.7)	99.8	0.98
BE19 SE	28 (1.7)	94.9	0.98
BE21 MN	24 (4.8)	105.1	0.97

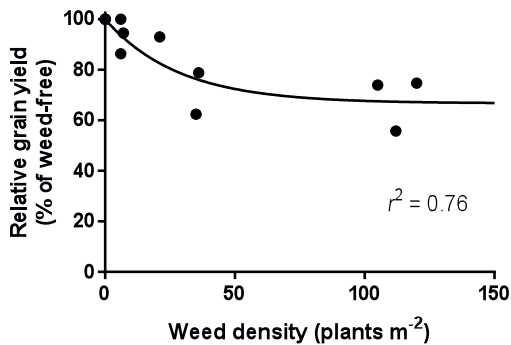
There was a hyperbolic relationship between bedstraw density and seed production ( $R^2 = 0.82-0.93$ ) in wheat and lentil (Figure 2). Despite delayed seedling emergence of bedstraw relative to both wheat and lentils, plants were still capable of significant seed production, which was considerably higher in lentils ( $>20,000$  seeds  $m^{-2}$ ) than wheat ( $3800$  seeds  $m^{-2}$ ) (Figure 2b). Seed production was also influenced by the growing season (Figure 2a), and was strongly correlated ( $R^2 = 0.78-0.96$ ) to rainfall and biomass production (data not presented). In response to above-average spring rainfall in 2016 (179 mm) at Roseworthy, bedstraw seed production in wheat was more than 1.5-fold higher (5900 seeds  $m^{-2}$ ) than in 2017 (3800 seeds  $m^{-2}$ ), which received below average spring rainfall (88 mm).

Bedstraw caused a reduction in wheat and lentil yield at low to moderate weed infestations (Figure 3). Maximum yield loss relative to the weed-free control for lentils ( $1937$  kg  $ha^{-1}$ ) was estimated to be more than 30%, whereas the losses were much smaller (10–14%) in wheat where weed-free yield in both growing seasons exceeded  $5$  t  $ha^{-1}$  (data not presented).

Even though bedstraw reached physiological maturity  $\sim 40$  d before the harvest-ready stage of both crops, it did not shed any seeds into the seed traps (100% retention, Table 2). The contamination of crop grain with bedstraw seeds was significantly higher in lentils ( $5092$  seeds  $kg^{-1}$  of grain) than wheat ( $12-234$  seeds  $kg^{-1}$  of grain). This result indicates that a large



**Figure 2.** Seed production of bedstraw in wheat in 2016 and 2017 growing season a), and in competition with wheat and lentils in 2017 b). The hyperbolic model was fitted in GraphPad Prism.



**Figure 3.** Relationship between density of bedstraw and relative grain yield of lentil (Hurricane XTA) in 2017. Relative yield = (weed density yield/weed free yield)  $\times$  100; Mean weed-free yield = 1937 kg ha<sup>-1</sup>. The non-linear curve represents a one-phase exponential decay model (GraphPad Prism).

**Table 2.** Bedstraw seed retention (%) in the inflorescence at the harvest-ready stage of wheat and lentils and grain contamination in 2016 and 2017. Values in parenthesis represent the SE of mean.

Crop (season)	Bedstraw seeds	
	Retention (%)	Grain contamination seeds kg <sup>-1</sup> of grain
Wheat 2016	100	234 (73)
Wheat 2017	100	12 (4)
Lentil 2017	100	5,092 (1,477)

amount of bedstraw seed ends up in the grain fraction during crop harvest and does not exit the harvester in the chaff fraction as is the case with weeds such as ryegrass.

## DISCUSSION

Seedling emergence behaviour was similar between the populations of bedstraw, however seedling recruitment was slow with most populations requiring 95–106 days to reach 50% emergence. In contrast, much faster rates of seedling recruitment (<30 days) have been observed for other common local weeds such as Indian hedge mustard and sowthistle, which are known to germinate rapidly and possess little or no innate dormancy (Chauhan 2006). Cumulative seedling emergence of populations ranged from 14–46% of the initial seedbank. Such high levels of ungerminated seedbank in bedstraw are consistent with previous reports of high seedbank persistence (Chauhan *et al.*

2006b). Seedling emergence in bedstraw populations only occurred after the onset of cold conditions in late winter (July–August), which is consistent with stimulation of seed germination in this species by cold-stratification (Chauhan *et al.* 2006a). Late seedling emergence observed in bedstraw would enable many plants to escape pre-sowing weed control tactics.

Despite its late seedling emergence relative to wheat and lentils, bedstraw was capable of large seed production, particularly in lentils where seed production exceeded 20,000 seeds m<sup>-2</sup>. Relative to wheat, lentils are known for their poor weed competitive ability (McDonald *et al.* 2007), and seed production of this magnitude may provide some explanation for the increasing prevalence of this weed in areas of lentil production. Even at moderate weed infestations (150 plants m<sup>-2</sup>), bedstraw reduced lentil grain yields by  $\geq$ 30%, whereas the yield losses in wheat were more modest (10–14%). Previously Rola (1971) reported that catchweed bedstraw was capable of reducing wheat yields by more than 30–60%, and attributed its high competitive ability to its climbing habit and ability to smother the crop.

In two growing seasons of contrasting spring rainfall, bedstraw did not shed any seeds into the seed traps (100% retained). This is in stark contrast to other weed species such as barley grass which shed more than 95% of its seed by crop harvest (Kleemann and Gill 2018). Complete seed retention in bedstraw was reflected in the high level of grain contamination with bedstraw seeds, especially in lentils (Table 2). The level of crop grain contamination was 25 to 400-fold higher in lentils than wheat, which could be partly related to greater weed seed production in lentils than wheat (Figure 2), but could also be associated with greater observed susceptibility of bedstraw to lodge in wheat. Catchweed bedstraw was also reported to easily lodge (Defelice 2002) because of its high biomass accumulation, weak stems, and tangled growth habit. In contrast to wheat, the short growth habit of lentils resulted in more vigorous bedstraw plants, which were less prone to lodging. Lower harvest height used in lentils than wheat (5 cm versus 15 cm) could have also contributed to greater grain contamination by bedstraw seeds in lentils. These results suggest that HWSC practices could assist in the management of bedstraw provided its seeds could be effectively captured and separated into the chaff fraction.

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