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Multi-year assessment of seed shedding for economically important grass weed species in Italy and the UK



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ABSTRACT

Harvest Weed Seed Control (HWSC) tactics aim to reduce weed dissemination and are considered promising approaches for future Integrated Weed Management (IWM) strategies. To be effective however, HWSC requires that target species have high seed retention at crop harvest. Here, a multi-year assessment of seed shedding was conducted across large geographical areas in the UK and Italy, for pernicious grass weed species that infest winter wheat and soybean crops. In the UK, an eight year assessment of Alopecurus myosuroides seed shedding was carried out in winter wheat crops. In Italy, seed shedding studies were conducted for three years, assessing A. myosuroides, Avena spp. and Lolium perenne ssp. multiflorum in winter wheat, and Sorghum halepense and Echinochloa crus-galli in soybean crops. Our results demonstrate low levels of seed retention (approximately 20 %) for A. myosuroides and Avena spp. at harvest, while higher mean seed retention (49 %) was found for L. perenne ssp. multiflorum. As such, Avena spp. and A. myosuroides are not good targets for HWSC across the studied locations, while HWSC could significantly contribute to L. perenne ssp. multiflorum management if combined with further control tactics. Seed retention at soybean harvest was on average 50 % for E. crus-galli, but higher at approximately 75 % for S. halepense. HWSC could therefore have a considerable impact on S. halepense populations in Italian soybean fields, but only an intermediate-low impact on E. crus-galli populations. Importantly however, we also find evidence for significant spatial and temporal variability in the extent of seed retention for all species. This study demonstrates that the potential for HWSC varies considerably between target weed species and highlights the importance of inter-annual variation in determining its expected performance.

1. Introduction

Due to their efficacy, ease of use, and cost-efficiency, herbicides have become the predominant method for weed control globally. Their widespread adoption has significantly eased the burden of weed management, providing farmers with both economic and operational benefits (Gianessi, 2013; Gianessi and Reigner, 2007). However, concerns have been raised about the environmental impact of excessive herbicide use, with contamination of ground and surface water bodies widely reported across European agricultural regions (Barchanska et al., 2017; Cruzeiro et al., 2015; Fingler et al., 2017; Kalogridi et al., 2014; Laini et al., 2012; Poulier et al., 2015). Recognizing a growing public demand for reductions in herbicide, and broader pesticide use, both the UK and the EU have committed to policies aimed at decreasing the agricultural reliance on these chemistries, including the UK "Environmental Land Management" scheme and the EU "Farm to fork" strategy (European Commission, 2020). These emphasize the importance of minimizing agrochemical inputs, encouraging the adoption of novel agroecological and technological approaches for weed management.

Overreliance on a small number of herbicidal modes of action (MOA) as the primary means of weed control has further led to the wholesale evolution and spread of herbicide resistant (HR) weed biotypes globally. Updated statistics on HR underline the magnitude and agronomic importance of this process: 533 unique cases (weed species x herbicide site of action) of HR weeds are currently reported worldwide, involving 273 species (156 dicots and 117 monocots) and affecting 21 of the 31 known herbicide sites of action for a total of 168 different herbicides (Heap, 2024). This issue affects both conventional and genetically or non-genetically modified herbicide tolerant cropping systems, and is worsened by the progressive simplification of crop rotation and weed management, and by the scarcity of new herbicidal MOAs. Three herbicides with innovative modes of action have been recently discovered

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(Umetsu and Shirai, 2020): cyclopyrimorate (site of action: homogentisate solanesyltransferase (HST), a downstream enzyme of HPPD), tetflupyrolimet (site of action: dihydroorotate dehydrogenase (DHODH), an enzyme connected with de novo pyrimidine biosynthesis), and cinmethylin (site of action: fatty acid thioesterase (FTA), leading to the inhibition of fatty acid biosynthesis); however, their commercialization across the EU and other world countries will take considerable time (Dayan, 2019). Moreover, weed populations with reduced sensitivity to some of these herbicides are already reported (Comont et al., 2024), emphasizing the need for careful management of these actives.

The threat of herbicide resistance and the need for careful stewardship of existing actives has necessitated the development of alternative, innovative tools and weed control strategies to reduce the reliance on herbicides (Liebman et al., 2016; Loddo et al., 2021; Owen, 2016). Targeting weed seeds in-crop is widely considered one of the most promising approaches for future sustainable weed management strategies (Shergill et al., 2020; Walsh et al., 2018b), particularly for weed species with short-lived non-persistent seeds such as many grass weeds, because their seed banks can be depleted quickly and effectively. This can be achieved either by targeting weed seeds within the crop cycle (usually called crop topping) or at harvest (Harvest Weed Seed Control). In the case of crop topping, mechanical, physical or chemical tools are used, alone or in combination, to kill weed plants within the cropping season, before seed ripening. For example, mowing/clipping of weed plants or herbicide top applications have shown promise for the control of Avena fatua L. (Tidemann et al., 2021) and Chenopodium album L. (Anderson et al., 2023). Specific machinery has been developed to pull weed plants overtopping soybean (Simard et al., 2019), while Moore et al. (2023) achieved a reduction of Amaranthus palmeri S. Watson seed production using an electrical treatment. Selectivity to crops is usually ensured thanks to differences in size (tall weeds in short crops) or phenology (early developing weeds in later developing crops).

Harvest Weed Seed Control (HWSC) in contrast, targets weed seeds at the point of crop harvest and can be implemented with a variety of tactics (Walsh et al., 2018b). Narrow-windrow burning involves channeling crop residues into a windrow that is then burned, and has proven effective in eliminating seeds of Lolium rigidum Gaudin and Raphanus raphanistrum L. in wheat residues (Walsh and Newman, 2007), and Lolium perenne L. ssp. multiflorum (Lam.) Husnot., A. palmeri, Sorghum halepense (L.) Pers., and Echinochloa crus-galli (L.) P. Beauv. in soybean residues (Norsworthy et al., 2020; Spoth et al., 2022). As an alternative strategy, combine harvesters can be equipped with specific mills, such as the "Harrington Seed Destructor" (Walsh et al., 2018a) and "Seed Terminator" (Winans et al., 2023), that destroy about 95 % of weed seeds entering the combine during harvest through physical grinding of the chaff. Furthermore, removing crop chaff, and the weed seeds within it, from fields with chaff carts or balers can also reduce the amount of seed entering the seedbank (Shirtliffe and Entz, 2005; Walsh and Powles, 2007). Chaff exiting the combine can be placed in narrow rows (chaff lining and chaff tram lining) that are subsequently used as tracks for tractor wheels and even sprayed with herbicides in order to minimize weed seed survival, germination and seedling establishment (Bennett et al., 2023; Walsh et al., 2021). HWSC was originally developed in Australia, where it is now successfully adopted across large areas of wheat production to control a range of weeds (Walsh et al., 2017; Walsh and Powles, 2022). It was subsequently introduced to the USA, where it is being developed for weed control in several crops (Maity et al., 2022; Norsworthy et al., 2016; Schwartz-Lazaro et al., 2017).

In the EU, grasses are the primary HR threat across the main cropping systems, with *Alopecurus myosuroides* Huds. (Keshtkar et al., 2015; Marshall et al., 2013; Petit et al., 2010), *Avena* spp. (Adamczewski et al., 2013; Papapanagiotou et al., 2012; Sousa-Ortega et al., 2023) and *Lolium* spp. (Scarabel et al., 2020; Torra et al., 2021) becoming a widespread problem in wheat production, while HR populations of *S. halepense* (Panozzo et al., 2017a; Papapanagiotou et al., 2022) and *E. crus-galli* (Löbmann et al., 2021; Panozzo et al., 2017b) are spreading

in maize and soybean fields in Central and Southern Europe. Implementing HWSC tactics to control these species could be an important means to diversify weed management, particularly in areas where environmental conditions strongly limit the range of alternative crops to cereals, such as Northern Europe or the Mediterranean. However, all HWSC techniques are effective only against species that retain the majority of seeds on the plants until crop harvest, and at or above the harvest height. For this reason, Walsh et al. (2018b) proposed to classify the potential HWSC efficacy in controlling a given weed species as low, intermediate, high and very high when seed retention as crop harvest was below 50 %, between 50 % and 75 %, between 75 % and 90 %, and above 90 %, respectively.

The assessment of weed-seed retention at crop maturity is therefore crucial to evaluate the potential benefit of introducing HWSC in a given cropping system. Large-scale studies have been undertaken to assess seed retention of the predominant weeds in US soybean (Schwartz-Lazaro et al., 2021a, 2021b) and wheat fields (San Martín et al., 2021; Soni et al., 2020), and spring wheat fields in Canada (Beckie et al., 2017; Burton et al., 2016, 2017). Although some studies have been conducted in Europe, e.g. (Akhter et al., 2020; Barroso et al., 2006; Bitarafan and Andreasen, 2020), inter-population and inter-annual differences in phenology and seed shattering dynamics (Necajeva et al., 2022) makes extrapolation from such studies at single sites or timepoints difficult. To provide a larger-scale evaluation of seed-shedding dynamics in economically important European grass weeds, here, a multi-year assessment of seed shedding was conducted across large geographical areas in the UK and Italy for pernicious grass weed species in winter wheat (A. myosuroides, Avena spp., L. perenne ssp. multiflorum) and soybean crops (E. crus-galli, S. halepense). The aim was to investigate geographical and inter-annual variation of seed shedding for the target species and estimate the potential for HWSC strategies within these cropping systems according to the classification proposed by Walsh et al. (2018b).

2. Materials and methods

2.1. Assessment of Alopecurus myosuroides seed shedding in the UK

The BGRI (Black-Grass Resistance Initiative) farm network was used to collect data on seed shedding in UK populations of *A. myosuroides*, growing in winter wheat crops. This farm network was set up in 2014 for evaluation of the distribution, abundance, and herbicide resistance of *A. myosuroides*, and covers the main winter cereal growing area of the UK. Initially the network had 73 farms with one or two fields per farm (Hicks et al., 2018). More fields were added in later years with two to four fields per farm, reaching approximately 200 fields in 2017. See Fig. 1 for locations of the surveyed fields.

Assessment of *A. myosuroides* seed shedding prior to harvest was carried out across this network in eight growing seasons (2014, 2017–2023). Assessments were typically executed over a six-week period leading up to harvest. Across this farm network, the number of winter wheat fields studied each year varies according to crop rotation. As such, the number of winter wheat fields visited was 124, 66, 61, 72, 22, 52, 71, and 81 in the years 2014, 2017, 2018, 2019, 2020, 2021, 2022 and 2023 respectively. Only 22 winter wheat fields were assessed in 2020, as the very wet autumn of 2019 limited autumn cereal sowing, resulting in a much reduced area of winter wheat cropping nationally. A total of 549 winter wheat fields were assessed over the 8 years.

Seed shedding was assessed at six random locations in each field; at least 10 inflorescences of *A. myosuroides* were checked to visually estimate the mean % of seeds shed at each location. The values of the six locations were then averaged to obtain an overall % seed shedding score per field. Each assessed field was visited once per growing season and the whole monitoring period lasted approximately 30–50 days. As a result, the phenological stage of both crops and *A. myosuroides* varied between the first and last assessed fields, representing a time-course of



Fig. 1. Maps to show the location of fields surveyed for weed seed shedding in (a) the UK, and (b) Italy. Points show the location of a single field. Crop type is shown by point colour, with blue for winter wheat, and red for soybean (Italy only). Fields were visited and seed shedding estimated over multiple years, UK: 2014 and 2017–2023, Italy: 2018–2020. Not all fields were visited every year.

seed shedding over this period each year. As such, an estimate of seedshed progression over this period up to harvest can be evaluated.

2.2. Statistical analysis of Alopecurus myosuroides seed shedding in the $U\!K$

The A. myosuroides percentage seed-shedding data from the UK was analysed using beta regression in R version 4.2.1. The response variable was the measured percentage of seed which had shed from the plant, while fixed effect terms were included for the effect of 'Julian day' (calendar days since 01st January), 'year', and their interaction. To better account for heteroscedasticity within the data, further terms were included to allow the precision parameter to vary with both 'Julian day' and 'year'. These variable dispersion beta regression models were fitted with a logit link, using the 'betareg()' function of the 'betareg' package (Cribari-Neto and Zeileis, 2010). To assess the significance of each predictor, a further series of nested models were fitted and compared using a likelihood ratio approach. To achieve this, separate sub-models were fitted excluding sequentially; the day*year interaction, the fixed effect of 'year', and the fixed effect of 'Julian day', and these nested models were assessed using the 'lrtest()' function of the 'lmtest' package (Zeileis and Hothorn, 2024).

To compare seed shedding against harvest progression, data for the UK winter wheat harvest were accessed from the Agriculture and Horticulture Development Board's (AHDB) survey of harvest progress. Specifically, data for the harvest progression of winter wheat crops was accessed, for the years 2017 – 2023. These data represent an annual, weekly measure of the percentage of winter wheat crops harvested nationally from the beginning of July to the end of September. Data for the 2014 harvest year were unavailable. To model harvest progression, a further variable dispersion logit-link beta regression model was fitted to this data, again using the 'betareg()' function of the 'betareg' package. In this case, fixed effect terms were included for the effect of 'Julian day', 'year', and their interaction, with the precision parameter allowed to vary with 'Julian day'. To gain an insight into the expected *A. myosuroides* seed remaining at harvest, the estimated mean and variance in seed-shed was predicted from the seed-shedding regression

model, over time-points representing the winter wheat harvest progression. Although not providing an exact quantification of seed-shed at harvest, this provides an estimation of the seed quantity likely to remain on heads at early and later points in the UK winter wheat harvest.

2.3. Assessment of grass weed seed shedding in Italian winter wheat and soybean fields

Seed shedding studies were conducted in Italy in the summer of 2018, 2019 and 2020. A. myosuroides, Avena spp. and L. perenne ssp. multiflorum were assessed in winter wheat fields during late-June to early-July, while S. halepense and E. crus-galli were studied in soybean from late-September to early-October. These species were selected due to their economic importance, being among the most problematic grass weeds for wheat and soybean production, and given the increasing occurrence of HR biotypes of these species in Italy (GIRE, 2025). All soybean fields surveyed were located in the region of Veneto, where approximately a third of Italy's soybean crops are grown. Winter wheat fields were assessed in four regions in 2018: Tuscany, Lazio, Veneto and Emilia Romagna. In 2019 and 2020, only winter wheat fields in Veneto and Emilia Romagna were visited (see Fig. 1). Fields visited were a mixture of farmers' fields, trial sites and research farms. Not all sites were visited every year since weed presence and density was not constant across years. In both crops, seed shedding was assessed only once per field 1-2 days before harvest.

Overall, 167 assessments were conducted on the 5 study species, with 33, 64, 22, 29 and 19 fields being assessed for *A. myosuroides, Avena* spp., *L. perenne* ssp. *multiflorum, S. halepense* and *E. crus-galli*, respectively. In some cases, the same field was used to monitor more than one species. The assessed fields were selected according to the presence of medium to large weed infestations, in order to provide enough weed individuals to be evaluated. Herbicide resistance/sensitivity status was not considered in site selection. To evaluate seed shedding, 100 - 200 individual inflorescences were assessed for each species at random points within each field. Inflorescences still without visible seeds, that is with BBCH index lower than 69–71, were not considered for the assessment since they could not produce viable seeds before crop

harvest. The level of seed shedding for each inflorescence was placed into one of five categories: Vlow (0 - 20 %), Low (21 - 40 %), Med (41 - 60 %), High (61 - 80 %), and Vhigh (81 - 100 %) according to a reference scale previously created for each species.

2.4. Statistical analysis of seed shedding in Italian grass weeds

The ordered categorical weed seed shedding data from Italy was analysed using a cumulative-link ordinal regression approach. Separate models were fitted for each individual weed species. The categorical weed seed-shedding data was modelled as the response variable, with 'year' as the fixed effect predictor. All models were fitted with a logit link, using the 'clm()' function of the 'ordinal' package (Christensen, 2023). For the species '*Avena*', data from 2018 were excluded as there were no observations in any seed shed category except "very high", leading to poor model convergence. Significance of the 'Year' term in each individual model was assessed from a type-I analysis of deviance using the Wald chi-square test, computed with the 'anova()' function. Additionally, comparison between individual years was conducted using a Tukey's post-hoc testing procedure, using the 'emmeans()' function of the 'emmeans' package (Lenth, 2024).

To evaluate the approximate quantity of seed remaining at harvest in each year, the percentage seed-shed scores from all inflorescences of a particular species were then averaged within each field, to produce an overall mean percentage of seeds shed in each species and field. The approximate percentage of seed retained on the plants at harvest was then estimated by subtracting the percentage seed shed from 100. The resultant values provide a distribution of estimated percentage seed remaining at harvest in each year, for each of the five weed species.

3. Results

3.1. Alopecurus myosuroides seed shedding in the UK

Seed shedding in UK populations of A. myosuroides began after approximately 160 Julian days (calendar days from 01st Jan), with the majority of seed shedding completed by approximately 210 - 230 Julian days. A significant 'day * year' interaction in analysis of the shedding data highlights that the rate of seed-shed progression varies from year to year (Table 1). Some years such as 2014 displayed a later and slower progression of seed shedding, while in other years (e.g. 2022) seed shedding began earlier and progressed more rapidly (Fig. 2). In contrast, the harvest of UK winter wheat crops began after approximately 200 Julian days at the earliest, progressing towards harvest completion at around 250 Julian days (Fig. 2). As with A. myosuroides seed shedding, the harvest progress also varied amongst years, resulting in a varying prediction of the amount of A. myosuroides seeds remaining on the plants at harvest from year to year (Fig. 3). In certain years (such as 2018), the relatively early wheat harvest led to a greater prediction of A. myosuroides seed retention at harvest in comparison to other years (Fig. 3). Nevertheless, in all years, only fields harvested relatively early in the UK wheat harvest period (approximately 0 - 30 % winter wheat harvest completion) are predicted to have any meaningful

Table 1

Statistical	analysis o	of UK Alc	pecurus r	nyosuroides	seed	shedding	data
			1	2			

Term	DF	Chi-sq	P-value
Day	1	482.196	< 0.001 * **
Year	7	312.168	< 0.001 * **
Day * Year	7	51.681	< 0.001 * **

Data was modelled using a variable dispersion beta-regression, with a logit link, and precision allowed to vary for both 'Day' and 'Year'. For each term and their interaction, Degrees of Freedom (DF), values of the Chi-squared test (Chi-sq) and statistical significance (P-value) determined from nested model comparisons using a likelihood-ratio test are reported.

A. myosuroides seed remaining on-plant at harvest.

3.2. Grass weed seed shedding studies in Italy in winter wheat and soybean fields

A significant effect of 'year' on seed shedding was detected for all studied grass species, but with different trends for weeds in wheat versus soybean crops (Table 2). Within each species, pairwise comparison of seed-shedding highlighted significant differences in seed-shed between all study years, with the exception of the comparison between years 2018 and 2020 in both *E. crus-galli and S. halepense* (Table 2). For *L. perenne* ssp. *multiflorum, A. myosuroides* and *Avena* spp. measured in wheat crops, the greatest level of seed shedding was observed in 2018. In particular in the case of *Avena* spp. only observations in the highest seed shedding category ('Vhigh', 81 - 100% seed shed) were recorded in 2018 (Fig. 4). In contrast, for *E. crus-galli* and *S. halepense* measured in soybean, slightly greater seed shedding was observed in 2019, albeit with relatively lower variation between years.

Greater variability in the level of seed shedding at harvest was observed amongst the grass-weed species assessed within Italian wheat fields (Fig. 4). As with the UK data, *A. myosuroides* seed heads at harvest were predominantly in the highest seed-shedding category (81 - 100 % of seeds shed) (Fig. 4). A similar pattern of relatively high seed shed at harvest was also observed for *Avena* spp. In contrast, lower levels of seed shedding were measured for *L. perenne* ssp. *multiflorum*, with a more even distribution of the inspected seed heads across the different shed categories - albeit with clear variability between sites and years also present. Consequently, the estimated proportion of seed remaining on plants at wheat harvest varied among these three species, with mean seed retention values of approximately 20 % for *A. myosuroides* and *Avena* spp. but higher mean seed retention at approximately 49 % in the case of *L. perenne* ssp. *multiflorum* (Fig. 5a).

Low to intermediate levels of seed shed were observed for *E. crus-galli* and *S. halepense* in soybean fields at harvest (Fig. 4b). In particular, most seed heads of *S. halepense* were on average classified in the 'Vlow' category (0–20 % shed), while the proportion of seeds shed from *E. crus-galli* was more variable across the sites and years of measurement. Consequently, higher seed retention of approximately 75 % was estimated for *S. halepense* at soybean harvest, with a mean seed retention of approximately 50 % observed for *E. crus-galli* (Fig. 5b).

4. Discussion

Harvest Weed Seed Control (HWSC) has shown considerable promise as a non-chemical means for managing a range of detrimental weed species across Australia (Walsh et al., 2017; Walsh and Powles, 2022) and parts of the USA (Maity et al., 2022; Norsworthy et al., 2016; Schwartz-Lazaro et al., 2017). To be effective however, this approach requires that the targeted weed species retain a high proportion of their seed on-plant at crop harvest. Prior to the introduction of HWSC tactics in the field, it is important therefore to examine the potential efficacy of this approach by quantification of seed retention amongst potential target species. Further complicating matters, differences in seed shedding for the same species across sites or between years have been widely reported for both grass and broadleaf species (Schwartz-Lazaro et al., 2022). As such, it is likely that multiple factors interact to determine the seed shedding dynamics and retention percentages at crop harvest. Here we demonstrate the extent of inter- and intra-specific variation in seed retention at harvest for a range of important European weed species.

4.1. Alopecurus myosuroides and Avena spp. are poor candidates for HWSC

In the current study, estimated seed retention for *A. myosuroides* in wheat fields at harvest in both Italy and the UK was lower than the values of 30–40 % reported by a previous study conducted in Denmark



Fig. 2. Alopecurus myosuroides seed shed versus winter wheat harvest in the UK. Percentage of *A. myosuroides* seed-shed measured in field (blue dots) is shown, while the lighter and darker blue shading represent the 95 % and 80 % quantiles for the estimated relationship for seed shedding over time. Red dots show the national average winter wheat harvest progress, with lighter and darker red shading representing the 95 % and 80 % quantiles for the estimated harvest progression. The grey shaded region depicts the 'harvest-window', i.e. the period between the start and end of winter wheat harvest in the UK in each year.

(Bitarafan and Andreasen, 2020). Although variation was observed from year to year, the majority of seed for this species is likely to have shed by the time of wheat harvest. The relatively higher seed retention reported by Bitarafan and Andreasen (2020) may relate to differences in crop and weed phenology associated with the geographical locations and climates in which those prior studies have taken place. Overall, the present study

clearly confirmed that in both the temperate UK climate, and warmer Italian conditions, *A. myosuroides* consistently shed seeds relatively early in comparison with other autumn-emerging grass weeds, as reported by Akhter et al. (2020). While HWSC may have had some potential for controlling seed return of *A. myosuroides* in early harvested UK wheat fields in specific years, overall this approach is predicted to have had a



Fig. 3. Estimated *A. myosuroides* seed remaining on the plant at harvest. The X-axis shows the progression through the UK winter wheat harvest period, as the proportion (%) of fields nationally which have been harvested. The fitted line shows the estimated proportion (%) of A. *myosuroides* seeds remaining on the plant, calculated from the fitted relationship for seed-shedding over time (shown in Fig. 2). The lighter and darker blue shading represent the 95 % and 80 % quantiles for the estimated relationship.

much lower impact in other years, and in the Italian study fields.

For Avena spp., 50–60 % of seed was predicted to be remaining at harvest in 2019 in some Italian wheat fields. However in other fields that year, and in almost all fields in 2018 and 2020, about 75 % of seed had already been shed. As a result, the overall estimated seed retention at harvest for Avena spp. was comparable with that of A. myosuroides, at

approximately 20 %. The limited seed retention observed for this species is consistent with the findings of several previous studies. Barroso et al. (2006) reported less than 10 % of seed retention at winter wheat harvest in Spain, as did Beckie et al. (2017) for spring wheat in Western Canada, while Tidemann et al. (2017) demonstrated that seed retention at spring wheat and faba-bean harvest in Western Canada greatly varied between

Table 2

Statistical analysis of seed shedding in separate weed species from Italy.

Model fitting				Post-hoc comparison				
Species	Term	DF	Chi-sq	P-value	Contrast	Estimate	SE	P-value
ALOMY	Year	2	156.72	< 0.001 * **	2018-2019	1.153	0.1081	< 0.001 * **
					2018-2020	1.590	0.1288	< 0.001 * **
					2019-2020	0.437	0.0856	< 0.001 * **
AVESP	Year	1	80.681	< 0.001 * **	2019-2020	-0.608	0.0677	< 0.001 * **
LOLMU	Year	2	284.95	< 0.001 * **	2018-2019	1.070	0.0635	< 0.001 * **
					2018-2020	0.541	0.0716	< 0.001 * **
					2019-2020	-0.530	0.0689	< 0.001 * **
ECHCG	Year	2	107.83	< 0.001 * **	2018-2019	-0.506	0.0758	< 0.001 * **
					2018-2020	0.174	0.0854	0.1034 ns
					2019-2020	0.680	0.0704	< 0.001 * **
SORHA	Year	2	9.447	< 0.01 * *	2018-2019	-0.17584	0.0724	0.040 *
					2018-2020	-0.00283	0.0759	0.999 ns
					2019-2020	0.17301	0.0646	0.020 *

Data was modelled using a cumulative link proportional odds model, with a logit link. In each case, significance of the 'Year' effect was assessed using a type-I analysis of deviance on the fitted model using a Wald Chi-square test. Degrees of freedom (DF), Chi-square test value (Chi-sq) and P-value are reported for each case. Post-hoc comparison between individual pairs of years (Contrast) was conducted using a Tukey's pairwise comparison. Estimate, Standard Error (SE) and Tukey's P-value are reported for each comparison.

sites and years, with an average of 30 %. However, Walsh and Powles (2014) observed almost 80 % seed retention at winter wheat maturity for *Avena fatua* in Western Australia, and similar values were also reported for spring wheat fields in Canada (Burton et al., 2016, 2017). Given this large variability in reported *Avena* seed retention at harvest, care must be taken before extrapolating findings more broadly to other crops or geographical areas. Taken together these results suggest that *Avena* spp. is unlikely to represent a 'good' target for HWSC, at least in the Italian wheat cropping regions evaluated here.

Given the limited impact estimated for HWSC on the control of both Avena spp. and A. myosuroides in the locations studied, in order to minimize seed return from plants which have escaped herbicide application or other control operations, efforts should be directed to design effective crop topping tactics to remove seed heads before seed ripening. Mowing/clipping of weed plants or herbicide top application have shown promise for the control of Avena fatua in Canada (Tidemann et al., 2021) These tactics are effective only if applied before seed ripening, so timing of control operations (e.g. early versus later clipping of heads) is crucial in determining seed production (Tidemann et al., 2020). Specific machinery has been developed to harvest seed heads of A. myosuroides from cereal fields before dissemination; (Top Cut Collect - Weed Harvester by Zürn Harvesting GmbH & Co. Kapellenstrasse 1, 74214, Schöntal-Westernhausen, Germany), however, limited adoption by farmers and the scarcity of experimental data makes the efficacy of this system difficult to currently assess.

4.2. Lolium perenne ssp. multiflorum, Echinochloa crus-galli and Sorghum halepense show greater potential for HWSC

On average, intermediate levels of seed retention (approximately 49 %) were estimated for L. perenne ssp. multiflorum, greater than that for the other grass weeds (A. myosuroides and Avena spp.) evaluated in wheat. Overall, seed retention for this species varied between approximately 27 % - 80 % over the sites and years studied. This is comparable with previous studies; San Martín et al. (2021) reported a range of 30-50 % of seed retention across several sites in the US Pacific Northwest, while Maity et el. (2022) also described large differences in seed shattering between different US regions, and finally extremely high level of seed retention (about 90 %) was observed in Kentucky (Herman and Legleiter, 2023). Due to this intermediate level of seed retention, HWSC tactics could potentially have a greater impact on populations of L. perenne ssp. multiflorum in Italian wheat fields than either A. myosuroides or Avena spp. Given the overall levels of seed retention observed, we may expect that at least 40-60 % of seeds produced every year could be controlled by HWSC tactics. This evaluation agrees with the estimates and direct observations reported in previous studies from different US regions (Beam et al., 2019; Herman and Legleiter, 2023; San Martín et al., 2021), confirming that HWSC could significantly contribute to *L. perenne* ssp. *multiflorum* control, if implemented within IWM strategies combining other chemical, mechanical, or cultural tactics.

In contrast to grass weeds in wheat crops, we observed generally higher and more consistent seed retention in the two weed species (E. crus-galli and S. halepense) assessed in soybean crops. In particular, we identified considerable (approximately 75 %) of S. halepense seeds retained on-plant at harvest in soybean, comparable with results reported from field trials in Texas (USA) by Schwartz-Lazaro et. al. (2021a). HWSC could therefore have a considerable impact on S. halepense populations in Italian soybean fields, as already suggested for the USA (Walsh et al., 2018b). However, HWSC has no effect on the vegetative propagation of this species, so further chemical or mechanical tactics would remain necessary. Nonetheless, minimizing seed dissemination by HWSC would significantly reduce dispersal ability of S. halepense, given that patches of plants originating from rhizomes are stable over time (Andújar et al., 2012). This would consequently enable site-specific management approaches such as patch spraying, reducing herbicide use required to control S. halepense. Additionally, limiting sexual reproduction and associated assortment and recombination of genotypes through seed destruction could help to slow the evolution of further weedy or resistance traits.

For E. crus-galli, overall mean seed retention was comparable with that of L. perenne ssp. multiflorum at 49 %, with values of seed retention for single sites ranging from 27 % to 64 %. These values are slightly lower than those described in previous studies conducted in the South-Central region of the US, where seed retention of E. crus-galli at soybean maturity was observed to range from 40 to above 80 % (Schwartz-Lazaro et al., 2017, 2021a). Such variability can be ascribed to the plasticity and genetic variability of E. crus-galli that enable this species to colonize areas with contrasting environmental conditions, with notable phenological differences observed amongst populations from different EU countries (Necajeva et al., 2022). It is worth noting however that E. crus-galli plants periodically produce new stems for as long as environmental conditions remain conducive to this, so seed heads at different levels of maturity are easily found on the same plant, further complicating seed shedding dynamics. Consequently, the implementation of HWSC tactics by Italian soybean growers may be expected to have an intermediate-low impact on E. crus-galli populations as already suggested for US soybean cropping areas (Schwartz-Lazaro et al., 2017, 2021a). It seems therefore reasonable to replace or combine HWSC with crop topping tactics that could control the early-developing



Fig. 4. Measurements of seed shed at harvest from weed species in (a) winter wheat, and (b) soybean fields in Italy. Seed shedding was recorded for 200 seed heads per field using a categorical scale; Vlow (0-20 % shed), Low (21-40 %), Med (41-60 %), High (61-80 %), and Vhigh (81-100 % shed). Bars show the mean (\pm Standard Deviation) proportion of seed heads in each category across all visited fields. Individual points show the values for each individual field visited, highlighting the extent of variability from one field to the next.



Fig. 5. Boxplots to show the estimated proportion of seed remaining at harvest in weeds of (a) winter wheat, and (b) soybean crops in Italy. Values were calculated from measurements of percentage seed-shed from 200 seed-heads per field, of each weed species (data shown in Fig. 4). Individual points show the mean percentage of seed remaining at each individual field-site visited.

individuals within the *E. crus-galli* populations. Given the large size differences between *E. crus-galli* and soybean, several crop topping tactics that have been proven effective against other weeds, e.g. herbicide wiper application for *A. palmeri* (Farr et al., 2022), mowing/trimming for *A. fatua* (Tidemann et al., 2021) or electrocution for *A. palmeri* (Moore et al., 2023), could be effectively adopted for *E. crus-galli* control. Similar tactics could also contribute to the control of *S. halepense* plants that often coexist in the same soybean field with *E. crus-galli*.

4.3. The importance of inter-annual variation

One consistent feature of all species evaluated was a significant variability in observed seed retention amongst sites and years studied. Weather conditions can play a key role in explaining annual or regional differences in seed shedding (Schwartz-Lazaro et al., 2021a) for *E. crus-galli.* Broadly, wetter and colder conditions are expected to extend weed growth and flowering stages leading to increased seed retention, while warm and dry conditions lead to earlier and higher seed shedding, as reported for *L. perenne* ssp. *multiflorum* (Maity et al., 2022), *Avena fatua* (Shirtliffe et al., 2000), but also Brassicaceae oilseed crops (Gan et al., 2008). Additionally, single 'intense' weather events could also determine seed shedding at a local level. Walsh and Powles (2014) reported that intense rainfall at harvest time significantly reduced seed retention for *A. fatua.* Intense winds can also promote seed shedding, particularly in the case of weed species whose seed heads stand above the crop canopy, as for *A. myosuroides* and *Avena* in wheat, and

E. crus-galli and S. halepense in soybean. Although not formally tested here, it is likely that wind could have contributed to the high level of seed shedding observed for Avena spp. and A. myosuroides in Italian wheat fields in 2018, given that the weeks before harvest were characterized by several windy days. Differences in intra- and inter-specific competition, such as with crops, could also influence seed shedding. Maity et al. (2022) reported density-dependent increases of seed shedding for L. perenne ssp. multiflorum, identifying that under high plant densities, and therefore higher competition, L. perenne ssp. multiflorum plants retained less seed at harvest. As a result, the observed interannual and site-to site variation in seed shedding within the current study are likely to stem from differences in weather, crop/weed competition, and field-level differences in agronomy amongst both the UK and Italian sites studied. Given that the combination of weather conditions, competition, and agronomy can all lead to considerable differences in seed shedding between years or sites for the same weed species, the relative impact of harvest weed seed control (HWSC) is also likely to vary considerably, making that the overall potential impact of HWSC in European arable systems difficult to predict.

5. Conclusions

Previously, a lack of knowledge about weed-seed retention at crop maturity for many important grass weed species in large UK and EU areas has prevented a thorough evaluation of the potential benefits of introducing HWSC. Here we have attempted to overcome this, providing

for the first time a multi-year assessment of seed shedding across large geographical areas in the UK and Italy, for pernicious grass weed species in winter wheat and soybean fields. We demonstrate that the potential for HWSC varies considerably according to target weed species: A. myosuroides and Avena spp. represent poor targets with only c.a. 20 % of seed remaining on the plants at harvest. L. perenne ssp. multiflorum (in wheat) and E. crus-galli (in soybean) are more promising for HWSC, with approximately 50 % seed retained, while S. halapense was found to retain approximately 75 % seed at soybean harvest, making this a good candidate for effective HWSC management. Nonetheless, the significant inter-annual variation observed in all species will affect the expected performance of HWSC strategies within these systems, making the overall efficacy of this approach less predictable than current herbicidedominated practices. Further studies are warranted to determine how local micro-climatic or weather conditions at the field scale can affect seed retention, driving the spatial and temporal differences observed. Thus, despite the fact that HWSC can significantly reduce seed return under some circumstances, its efficacy is likely to vary considerably amongst species, sites, and years, which may be problematic for its effective implementation for the control of the studied species in European cropping systems.

CRediT authorship contribution statement

Loddo Donato: Writing – original draft, Methodology, Investigation, Data curation, Conceptualization. Hull Richard: Writing – review & editing, Supervision, Project administration, Methodology, Investigation, Funding acquisition, Conceptualization. Sattin Maurizio: Writing – review & editing, Funding acquisition, Conceptualization. Comont David: Writing – original draft, Visualization, Software, Funding acquisition, Formal analysis, Data curation.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data Availability

Data will be made available on request.

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