Weed Seed Fate during Summer Fallow: The Importance of Seed Predation and Seed Burial

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Maximizing weed seed exposure to seed predators by delaying post-harvest tillage has been suggested as a way to increase weed seed loss to predation in arable fields. However, in some areas of northeastern Spain, fields are still tilled promptly after cereal harvest. Tillage usually places seeds in a safer environment compared to the soil surface, but it can also increase seed mortality through seed decay and fatal germination. By burying the seeds, tillage also prevents weed seed predation. Weed seed fate in a tilled vs. a no-till environment was investigated during the summer fallow months in three cereal fields in semi-arid northeastern Spain. Rigid ryegrass and catchweed bedstraw seeds were used. Predation rates were measured in a no-till area within each field in 48-h periods every 3 wk, and long-term predation rates were estimated. Fate of buried seeds was measured by burying 20 nylon bags with 30 seeds of each weed species from July to September at a depth of 6 cm in a tilled area contiguous to the no-till area. Predation rates over the entire summer were 62% and 49% for rigid ryegrass and catchweed bedstraw, respectively. High availability of crop seeds (preferred by ants) on the soil surface may have decreased predation of weed seeds early in the season. Seed loss due to burial was 54% and 33% for rigid ryegrass and catchweed bedstraw, respectively. Unusual above-average precipitation probably prompted higher than normal weed germination rates (fatal germination) in some fields, and thus led to higher seed mortality rates compared with an average year. These results suggest that leaving the fields untilled after harvest may be the optimum strategy to reduce inputs to the weed seedbank during the summer fallow period in semi-arid systems.

Nomenclature: Catchweed bedstraw, *Galium aparine* L; Rigid ryegrass, *Lolium rigidum* Gaudin.

Key words: Background seed density, crop seeds, harvester ants, seed preference, semi-arid, tillage.

Postdispersal weed seed predation is an important ecosystem service that can help reduce the number of weed seeds entering the seedbank after being dispersed from the mother plant (Westerman et al. 2006, 2012). To maximize seed loss due to predation, some studies have suggested increasing exposure time of weed seeds to predators by allowing them to remain on the soil surface after being shed (Westerman et al. 2006, 2009). In annual arable crops such as winter cereals, this can be achieved by delaying some management practices like harvest or soil disturbance, mainly tillage after harvest. Opportunities for delaying tillage can be difficult to find within a cropping system, since there may be other reasons for soil tillage that may offset seed predation termination. However, in other circumstances, tillage could be delayed without many drawbacks. In semi-arid regions such as the rainfed areas of northeastern Spain, low rainfall allows just one crop per year, mainly winter cereals, which grow from late October–early November to late June–early July. After crop harvest, fields are left fallow until the next sowing season in autumn. Despite widespread adoption of no-till practices in certain semi-arid areas, some farmers still chisel plow the fields after cereal harvest. Reasons for tillage include burying the crop stubble, preventing soil compaction in autumn, managing summer weeds, and burying winter weed seeds.

Stubble tillage may have contrasting effects on weed populations. Usually, tillage places seeds in a “safer” environment, the soil matrix, compared with the soil surface (Mohler 1993, and references therein). In this sense, tillage can contribute to the buildup of the weed seedbank (Buhler et al. 1997). However, tillage can also be used as a weed management strategy if seeds are buried to a depth from which they cannot successfully emerge (fatal germination) (Chauhan et al. 2006; Cousens and Moss 1990; du Croix Sissons et al. 2000) or if they are attacked by soil microorganisms (seed decay) (Davis et al. 2006; Gomez et al. 2013; Pekrun and Claupein 2006). In semi-arid systems, conservation tillage and no-till seem...
to enhance soil microbial biomass carbon and soil enzymes, which are indicators of microbial activity, compared with conventional fully tilled systems (Alvaro-Fuentes et al. 2013). However, there is still very little information about the effects of those organisms on weed seeds, especially during the summer.

In rainfed cereal fields in NE Spain, harvester ant (Messor barbarus L.) populations are large, especially in no-till and minimally tilled fields (Baraibar et al. 2009, 2011a). Ants are active from spring to late autumn, and their activity coincides with the period of seed shed and seed availability on the soil surface for most of the important weeds in these systems. Seed removal by harvester ants was estimated to be around 72% of the weed seeds produced annually before crop harvest (Westerman et al. 2012). So, seed predation in these systems seems to be an effective strategy to decrease weed seed incorporation into the seedbank. Because it buries seeds, tillage stops weed seed predation, since ants and most seed predators do not dig for buried seeds.

Though preharvest seed predation has been studied, postharvest predation has seldom been addressed (but see Baraibar et al. 2009; Spafford Jacob et al. 2006), and many questions still remain regarding weed seed fate during this period. For example, seeds could be buried under the straw after cereal harvest and become less visible for ants, thus decreasing seed encounter rates by predators and, consequently, removal rates (Baraibar et al. 2011b; Westerman et al. 2006, 2009). Similarly, the density of different seed species on the soil surface after harvest (referred as seed environment or background seed density) may influence predation rates of different weed species, since it can change ants’ seed preference (Detrain et al. 2000; Lopez et al. 1993; Reyes-Lopez and Fernández-Haeger 2002; Risch and Carroll 1986). Seed environment on the soil surface can also change ants’ spatial exploration range (Detrain et al. 2000; Lopez et al. 1993). If resources are scarce, ants may explore a wider area of the field, whereas if seeds are available in high densities close to the nest, the area explored may be smaller and some seeds placed away from the nest may remain undiscovered and eventually enter the seedbank. Understanding seed predation magnitude after crop harvest and factors influencing it can help design management practices to try to maximize it.

In this study, we followed weed seed fate in a tilled vs. a no-till area in three cereal fields during the summer fallow period. In the no-till areas, we measured seed predation rates of two weed species (presented in petri dishes) and assessed the effects of straw on seed encounter and seed predation rates. We also measured seed background density to understand whether the seed environment experienced by predators during the summer influenced predation rates of our experimentally added seeds or ants’ exploration behavior. In the tilled areas, we followed the postburial fate of a known number of seeds. Our objective was to compare mortality rates of exposed vs. buried weed seeds during the summer fallow to inform our recommendations to farmers about weed seed management strategies during this period.

Materials and Methods

Site. Experiments took place in the rainfed cereal region of northeastern Spain. Mean annual temperature is 15 C, and average rainfall is 342 mm (Agencia Estatal de Meteorología 1983–2010), concentrated mainly in spring and autumn. Summers are hot and dry (average maximum: 31 C), and winters are mild (average minimum: 2 C). Accumulated rainfall at the end of the summer of the experiment (2014) was between 31% and 118% higher than the average (120 to 214 mm more than average depending on the location). Three winter cereal fields were chosen, one in Algerri (41.82° N, 0.59°E), one in Balaguer (41.80°N, 0.76°E), and one in Vilanova de Bellpuig (41.59°N, 0.98°E). The field in Algerri was organically certified and followed a 3-yr rotation of wheat (Triticum aestivum L.), spelt (Triticum spelta L.), and a mixture of oats (Avena sativa L.) and vetch (Vicia sativa L.) for forage. The other two fields were farmed conventionally (not organically) and grew barley (Hordeum vulgare L.) as a monoculture. Algerri was the northernmost field, 100 m in altitude higher than the other two locations, and received considerably more rainfall than the other two. Based on climatology and the farmer’s schedule, cropping operations such as planting or tillage in Algerri occurred 2 wk later than in the other two fields. The three fields were chisel plowed at a depth of 15 to 20 cm two times a year, once after crop harvest in June/July and again before crop sowing in October.

Experimental Design. Per field, two 50 by 50 m areas were flagged. The two areas were separated by a 10- to 20-m strip. Two or three days after harvest, while the straw was still on the fields, seed predation was measured for 48 h in one of the areas (called the “no-till area,” see Weed Seed Predation section for experiment details). After this first seed predation
assessments took place, the straw was removed and the entire field was chisel plowed, except for the “no-till area” and the contiguous strip, which were left un-tilled throughout the summer.

Weed Seed Fate in the No-Till Area: Weed Seed Predation. The no-till area was used to measure seed predation during summer. Two weed species were used, rigid ryegrass (1.8 g per 1000 seeds) and catchweed bedstraw (6.8 g per 1000 seeds). These species were chosen because they differ in weight, size, shape, and preference by predators. Rigid ryegrass is highly preferred by harvester ants over catchweed bedstraw if the two are offered together (Westerman et al. 2012). Rigid ryegrass seeds collected from areas close to the experiments were used for the first seed predation measurement date, and seeds from Herbiseed (Reading, UK) were used for later dates. All catchweed bedstraw seeds were collected from local populations close to the fields.

To measure weed seed predation, 25 petri dishes (9-cm diameter) per species were installed in the “no-till area” of each field (50 petri dishes total). Each dish had two 15-mm-wide openings in the sides to facilitate predator access. Seeds of the two species were not mixed; each species had its own petri dish. Petri dishes from the same species were arranged 10 m apart on two regular grids of 5 rows and 5 columns. Petri dish grids from the two species were 5 m apart from each other (Figure 1). Seed removal was measured throughout the summer every 3 wk from harvest in June until September (Table 1). Seed removal in Algerri was measured four times during summer, while seed removal was measured five times in the other two fields.

On the first measurement date, 2 g of each species were left on each of the dishes to resemble high weed seed densities immediately after harvest. Seeds were left in the field for 48 h and then retrieved and weighed in the lab. Seeds returning from the field were dried before being weighed to ensure similar moisture contents before and after field exposure. The difference between the initial and the recovered weight was considered to represent seeds removed by predators. On this first date, right after cereal harvest, fields still had straw arranged in lines approximately every 10 to 15 m, covering around 20% of the field area. Accordingly, 20% of the petri dishes (10 random dishes per species) were placed under the straw lines.

Table 1. Harvest date, bag burial and recovery dates and date when weed seeds were placed on each field to estimate predation rates.

| Field          | Harvest date | Bag burial and recovery dates | Seed placement dates to estimate predationb |
|----------------|--------------|--------------------------------|-----------------------------------------------|
| Vilanova de Bellpuig | June 5, 2014 | July 22, 2014                   | June 6, 2014                                   |
|                |              | September 29, 2014              | July 8, 2014                                   |
|                |              | (68 d)a                         | July 29, 2014                                   |
|                |              |                                 | August 25, 2014                                |
|                |              |                                 | September 18, 2014                             |
| Balaguer       | June 7, 2014 | July 4, 2014                    | June 11, 2014                                   |
|                |              | October 6, 2014                 | July 8, 2014                                    |
|                |              | (93 d)a                         | July 29, 2014                                   |
|                |              |                                 | August 25, 2014                                |
|                |              |                                 | September 18, 2014                             |
| Algerri        | June 21, 2014| July 28, 2014                   | July 8, 2014                                   |
|                |              | September 19, 2014              | July 29, 2014                                   |
|                |              | (54 d)a                         | August 25, 2014                                |

a Recovery date. Number in parentheses indicates the number of days seeds were buried.
b Seeds were retrieved after 48 h.
with the distance between dishes kept constant as much as possible. The straw layer was approximately 15-cm thick. Straw was removed by farmers 2 d after the first measurement date. On the subsequent measurement dates, 1 g of each species was used. Field days were chosen to avoid extreme weather conditions such as strong wind and heavy rain so seed weight removed could be attributed to seed predators and not to climatic conditions.

**Seed Background Measurements.** The same day that dishes were placed for the predation assessment, background seed density was measured in five random 1-m² quadrats using an insect suction sampler (Vortis insect suction sampler, Burkard Manufacturing, Rickmansworth, UK). Sampled areas were noted to avoid resampling of the same area on later dates. Samples were taken to the lab, and seeds were separated from dirt and straw using a set of sieves. Seeds were identified to species and counted. Seed background measurements were correlated to preation and dish encounter rates (see Ant Searching Activity section).

**Harvester Ant Nest Density.** In August, all harvester ant nests in the no-till area were counted to get an estimate of predator density in each field. Once a nest had been counted, the area next to the entrance was sprayed with colored paint to prevent double counting. Nest density per hectare was calculated.

**Ant Searching Activity.** Ant searching activity was approximated by calculating the proportion of petri dishes found by ants in each sampling date. Dishes were considered found if the proportion of seeds removed was higher than 20% of the initial seed weight (following Baraibar et al. 2011a).

**Weed Seed Fate in the Tilled Area.** In the tilled area, 20 nylon bags per species (10 by 12 cm and 0.08-cm mesh size) were buried 6-cm deep. This depth was chosen because it is the depth at which most seeds are buried after chisel cultivation (Mohler et al. 2006). Each bag contained 30 seeds of one of the species together with 200 cm³ of soil. The goal of soil incorporation was to decrease the possibility of a fungal contamination from seed to seed (Van Mourik et al. 2005) and mimic real burial conditions, in which seeds are usually mixed up with soil particles. Burial location was random within the 50 by 50 m area. Bags were left buried throughout summer and were retrieved at the end of September or beginning of October (Table 1) based on the farmers’ intentions to till the fields. Bags were taken to the lab and frozen until they could be processed. Bag contents were washed using an elutriator (Wiles et al. 1996), and seeds were recovered using sieves and visual inspection. Recovered seeds were classified as either dead or alive. Dead seeds included those that had germinated (if we could still see the hypocotyl or the radicle), were decomposed, or were nonviable (no visible signs of germination, dead embryo). Decomposed seeds could have decomposed after germination, but we could not assess the causes of mortality for this group (10% of the seeds). Nonviable and live seeds were separated by incubating them in a 1% triphenyl-tetrazolium chloride for 48 h. Initial seed viability used to correct the results of viable seeds was 100% for rigid ryegrass and 96% for catchweed bedstraw.

**Statistical Analyses**

*Weed Seed Fate in the No-Till area: Weed Seed Predation*. A Bayesian analysis framework with Markov Chain Monte Carlo (MCMC) techniques was adopted to estimate the proportion of seeds lost to predators. Bayesian inference is a method of statistical inference in which Bayes’s theorem is used to update the initial probability for a hypothesis (prior) as more evidence or information becomes available. Because the dependent variable was a ratio, Bayesian methods seemed an appropriate approach to analyze the data. First, data exploration was applied following the protocol described in Zuur et al. (2010) to investigate outliers, homogeneity, normality, zero trouble, collinearity, relationships, interactions, and independence. Then, a beta distribution was used to model the ratio of predated weight and initial weight. This distribution is appropriate if the response variable is a continuous variable ranging from \( x_1 \) to \( x_2 \), with a logistic link function (Zuur and Ieno 2016). To account for the repeated measurements on the same petri dish, a random intercept “station” term was added. Hence, the following PROC GLMM was used:

\[
\text{Ratio}_{ij} \sim \beta (\theta \ast \Pi_{ij}, \theta \ast (1 - \Pi_{ij}))
\]

\[
\text{Expected} (\text{Ratio}_{ij}) = \Pi_{ij}
\]

\[
\text{Logit} (\Pi_{ij}) = \text{Date}_{ij} \ast \text{Weed}_{ij} + \text{Background}_{ij} + \text{Field}_{ij} + \text{Station}_i
\]

where \( \text{Ratio}_{ij} \) is the \( j^{th} \) observation at station \( i \), and \( \text{Station}_i \) is a random intercept that is normally distributed.

\( \text{Date}_{ij} \) is a random intercept that is normally distributed.
distributed with mean 0 and variance $\sigma^2$. Date, weed species, and field were fit as categorical variables. Field was treated as a fixed effect because of too few levels of the factor (Bayesian statistics require at least five levels to be able to consider an effect to be random, AF Zuur, personal communication). MCMC techniques were then applied to estimate the parameters in the model. Analyses were done with a program for Bayesian hierarchical models called JAGS (Plummer et al. 2016) via the package ‘R2jags’ in R (R Core Team 2014). Diffuse normal priors were used. A prior is the probability distribution of the initial hypothesis. Because previous knowledge about the prior distribution was lacking, a normal distribution with large variance (diffuse) was used. A half-Cauchy (5) distribution was used for $\sigma$, and a half-Cauchy (25) was used for theta. Sigma and theta are shape parameters of the beta distribution. Three chains were used, each with a burn-in of 25,000 iterations (burn-in refers to the practice of discarding an initial portion of a Markov chain sample so that the effect of initial values on the posterior inference is minimized). The thinning rate was 10 (i.e., every 10th draw from the algorithm is actually used to compute credible sets and medians of the posterior distribution), and 15,000 iterations were used for the posterior distribution of each parameter. Once the model was fit, a model validation was applied to ensure normality and homoscedasticity of residuals. Estimated proportions of seed loss were later used to estimate long-term loss due to predation.

Long-Term Loss Due to Predation. Long-term loss due to predation was calculated in two separate ways. Both methods followed the models by Westerman et al. (2003) and Davis et al. (2011), but they differed on the seed availability used as the basis for the calculations. Models used by Westerman et al. (2003) and Davis et al. (2011) were developed to estimate the annual proportion of newly produced weed seeds consumed by granivores $\overline{Mp}$ before crop harvest. In these models,

$$\overline{Mp}=\frac{1}{n}\sum_{i=1}^{n} \left( Y_i \prod_{j \neq j} S_j \right)$$

where $\overline{Sp}$ is the annual proportion of newly produced seeds that survive predation and can be calculated as:

$$\overline{Sp}=\frac{1}{n}\sum_{i=1}^{n} \left( Y_i \prod_{j \neq j} S_j \right)$$

These equations are used to estimate weed seed loss during weed seed rain. Consequently, annual seed production, $Y$, is divided into multiple pulses ($Y$) occurring within $n$ periods of a specified length. Seeds from each pulse survive at the episodic rate $S$, where the $k$ is the duration of the seed exposure. In our experiment, there was no seed rain. Weed seed rain stopped with cereal harvest, so the only seeds available to predators were the ones present on the soil surface on the first sampling date (first measurement of background seed density). However, densities of seeds on the soil surface were so unexpectedly low that the model estimated loss to predation to be 100% at 4 d after the start of the experiment. This was not supported by our background measurements, which showed rigid ryegrass seeds even on the last sampling date. So, we calculated long-term loss following Equations 2 and 3, assuming that seeds offered in petri dishes (g) plus the background seed weight of rigid ryegrass and catchweed bedstraw on each sampling date constituted the seeds available in each date.

**Results and Discussion**

**Weed Seed Fate in the No-Till Area.** The number of alive vs. dead seeds was analyzed using a GLMM with a binomial error distribution and a linear mixed model, respectively. Fields and petri dishes were treated as random effects, while bag number was entered as a fixed effect. Here, field was considered a fixed effect to enable comparison of mortality rates due to burial with mortality caused by predation for each field.

Straw Effect on Number of Dishes Encountered and Seeds Removed. The effects of the straw on the number of dishes found and the weight of seeds removed were analyzed using a GLMM with a binomial error distribution and a linear mixed model, respectively. Fields and petri dishes were treated as random effects, while weed species and the presence/absence of straw as fixed effects. Seed weight was log($x + 1$) transformed before running the analysis. All analyses were performed using the ‘lme’ package in R (R Core Team 2014).

Weed Seed Fate in the Tilled Area. The number of
Ieno (2016). The posterior mean of the fitted values and 95% credible intervals per field and weed combination for all dates are presented. Posterior means are the predicted values for seed predation rates, which are conditional on the observed data. There are no P-values in Bayesian statistics, and significance of the factors cannot be determined from the table. However, since we were not comparing different treatments, the significance of each factor is not as important as the overall trend of removal rates. Table 2 provides the numerical output to estimate weed seed predation rates for each combination of weed, field, and date. Interpreting this numerical output can be challenging (Zuur and Ieno 2016), so the fitted values are sketched in Figure 2 (minor random jittering was applied along the x-axis to ensure that points are not superimposed). The proportion of seed loss due to predation ranged from 20% to 80% per 2 d (Figure 2). Main seed predators were probably harvester ants, which were present in high densities in those fields (340, 335, and 360 nests ha⁻¹ in Vilanova de Bellpuig, Balaguer, and Algerri, respectively). Granivorous mice are very scarce in the area and leave clear signs when they visit petri dishes that were not seen in this experiment (i.e., droppings, seed chaff; Baraibar et al. 2009). Thus, rodents likely did not contribute to seed removal. We cannot exclude the possibility that birds removed some seeds. However, the few studies that have investigated seed loss due to bird predation show that such loss tends to be low during the summer (Holmes and Froud-Williams 2005).

Predation rates were low in June and tended to increase after the beginning of July. Weed seed predation rates observed were in accordance with other studies in the region for the same time period (Baraibar et al. 2009), thus suggesting that seed removal patterns by harvester ants in this region are consistent over time. Low removal rates immediately following crop harvest had already been documented by Baraibar et al. (2009). One of the possible causes for these low rates is background seed densities. Table 3 shows the density of crop and weed seeds recorded on each sampling date. Weed seed density was extremely low, and most of the seeds present on the soil surface were crop seeds (barley or spelt grains). A significant negative relationship between background density and seed predation of our experimentally added seeds was observed in Algerri and Balaguer (R² = 0.54 and 0.6, P = 0.02 and 0.015, respectively). This relationship suggests that when crop seeds were abundant (i.e., right after harvest), harvester ants consumed them preferentially over weed seeds and especially avoided less preferred seeds like catchweed bedstraw (Crist and MacMahon 1992). Then, as crop seed density decreased, ants consumed more of the experimentally added weed seeds. Similarly, we observed in these two fields a negative correlation between background seed density and the proportion of dishes found, although it was only significant in Balaguer (R² = 0.27 and 0.70, P = 0.18 and 0.002, respectively), suggesting that ants foraged closer to the nest when resources were high (i.e., right after

| Estimated mean | SE  | 2.5%  | 97.5% |
|----------------|-----|-------|-------|
| (Intercept)    | -0.39 | 0.23  | -0.84 | 0.05  |
| Date 2         | -0.67 | 0.25  | -1.16 | -0.18 |
| Date 3         | 0.10  | 0.26  | -0.40 | 0.62  |
| Date 4         | 0.59  | 0.25  | 0.10  | 1.09  |
| Date 5         | 1.61  | 0.26  | 1.09  | 2.13  |
| L. rigidum     | 0.16  | 0.27  | -0.37 | 0.70  |
| Field Balaguer | 0.28  | 0.13  | 0.04  | 0.53  |
| Field Vilanova de Bellpuig | -0.11 | 0.13 | -0.36 | 0.15 |
| Background     | 0.20  | 0.06  | 0.08  | 0.31  |
| Date 2 * L. rigidum | 0.88 | 0.35 | 0.19  | 1.56  |
| Date 3 * L. rigidum | 0.67 | 0.35 | -0.02 | 1.37  |
| Date 4 * L. rigidum | 0.39 | 0.35 | -0.30 | 1.07  |
| Date 5 * L. rigidum | -0.12 | 0.34 | -0.79 | 0.55  |
| Sigma²        | 0.06  | 0.05  | 0.01  | 0.17  |
| Theta²        | 0.43  | 0.02  | 0.40  | 0.47  |

* Sigma and theta are shape parameters of the beta distribution.
harvest) and shifted to search a wider area as resources became scarcer. This fact is in agreement with other studies that showed that harvester ant foraging trails are shorter and more branched when seed availability is high (Lopez et al. 1993). In the third field (Vilanova de Bellpuig), which had the highest background seed density (Table 3), weed seed removal was not significantly correlated to background seed density or to the proportion of dishes found. The lack of correlation was mainly driven by what happened at the first sampling date, when seed availability on the soil surface was highest. In contrast to the other fields, the small chaff portion of the straw (together with weed and crop seeds) was dumped from the combine and left in several piles across this field during harvest. An average of up to 217 crop seeds and 25 rigid ryegrass seeds were counted in a 10-cm³ volume taken from

Table 3. Background seed density: crop, rigid ryegrass, and total seed density (seeds m⁻²) in each field and sampling date.

| Field name       | Date           | Crop seeds m⁻² | Rigid ryegrass seeds m⁻² | Total seeds m⁻² |
|------------------|----------------|----------------|--------------------------|-----------------|
| Vilanova de Bellpuig | June 6, 2014   | 77.8           | 18.4                     | 96.2            |
|                  | July 8, 2014   | 6.8            | 0                         | 6.8             |
|                  | July 31, 2014  | 45.8           | 3.6                      | 49.4            |
|                  | August 25, 2014| 4              | 0                         | 4               |
|                  | September 18, 2014| 13.4      | 13.2                      | 26.6            |
| Balaguer         | June 11, 2014  | 17.6           | 0                         | 17.6            |
|                  | July 8, 2014   | 20.6           | 0                         | 20.6            |
|                  | July 29, 2014  | 3.2            | 0                         | 3.2             |
|                  | August 25, 2014| 9              | 1.4                      | 10.4            |
|                  | September 18, 2014| 0           | 0                         | 0               |
| Algerri          | July 8, 2014   | 69.6           | 2.8                      | 72.4            |
|                  | July 29, 2014  | 18.8           | 2.6                      | 21.4            |
|                  | August 25, 2014| 3              | 1.6                      | 4.6             |
|                  | September 18, 2014| 0.4       | 5.2                      | 5.6             |
these mounds (unpublished data). Those high-density areas did not occur in the two other localities and may have caused a change in ants’ foraging behavior (Detrain et al. 2000; Lopez et al. 1993). Ant trails consistently ended in the high-density piles, probably because this foraging strategy maximized resource acquisition (Detrain et al. 2000). The proportion of dishes encountered on this date and in this field was close to 100%, thus confirming that a larger area was explored but removal of weed seeds from the dishes was low, especially for the less preferred catchweed bedstraw seeds. These results suggest that crop seeds were "distracting" ants from eating weed seeds; thus, weed seed predation rates might have been higher if the density of crop seeds on the soil surface had been lower. Avoiding cleaning out harvesting machinery in the field or careful adjustment of combines to prevent crop seed loss at the time of harvest may be two strategies to reduce the density of crop seeds on the soil surface and increase weed seed removal. Similarly, the use of machinery designed to grind weed seeds as they come out of the combine (e.g., Harrington Seed Destructor; Jacobs and Kingwell 2016) could also decrease the amount of weed and crop seed returning to the soil surface after harvest and prompt ants to search a wide area of the field and be less selective concerning the weed seeds they harvest.

Contrary to our expectations, straw did not seem to be an obstacle for ants in locating or exploiting weed seeds located underneath the straw on the first sampling date. Straw did not seem to be a barrier to ant movement and did not prevent foraging or dish encountering. This result is encouraging, because it means that seed loss to predation right after crop harvest is not being limited by straw, and straw management by farmers therefore does not require any changes (Spafford Jacob et al. 2006).

**Long-Term Seed-Removal Rates.** Long-term seed loss to predators calculated following Equations 2 and 3 was 60%, 65%, and 62% for rigid ryegrass and 47%, 55%, and 44% for catchweed bedstraw in Vilanova de Bellpuig, Balaguer, and Algerri, respectively. Westerman et al. (2012) reported that 25% to 40% of rigid ryegrass seeds are shed at the time of harvest; thus, predation rates during the fallow period such as those reported here are likely to have a large impact on populations of this species. Loss may be even higher if combined with predation rate before harvest (0.67 for rigid ryegrass; Westerman et al. 2012). There is less information about seed-shed timing of catchweed bedstraw in semi-arid systems. A close relative, false cleavers (*Galium spurium* L.), sheds most of its seeds before harvest and has high levels of predation before the summer fallow (Westerman et al. 2012). Removal rates during the summer months could help further decrease the number of seeds of this species entering the seedbank.

**Seed Fate in the Tilled Area.** The interaction of the effects of weed species and field on seed mortality in the soil was significant. Proportions of dead seeds are shown per field and weed species separately in Figure 3. Seed mortality due to burial was highly variable across the fields and ranged from 52% to 58% for rigid ryegrass and from 24% to 50% for catchweed bedstraw. Rigid ryegrass seed mortality was significantly greater than for catchweed bedstraw seeds.
except in Vilanova de Bellpuig. Rigid ryegrass seed loss caused by burial was significantly higher in Vilanova de Bellpuig and Algerri compared with Balaguer, whereas catchweed bedstraw mortality was significantly higher in Vilanova de Bellpuig compared with the other two fields. Main causes of mortality also differed across fields. Whereas germination was the most important cause of weed seed loss in Vilanova de Bellpuig, in the other fields, and especially in Balaguer, seed decay was the main cause of mortality. Differences in causes of mortality were unexpected and remain unexplained, as we lack information about soil properties and microbial activity in those fields. Future research should explore seed mortality due to decay during summer and the main factors driving it, since decay can be a major factor in weed seed mortality.

Seed mortality due to burial (germination plus decay) was unexpectedly high considering the relatively short period of time the seeds were buried (Boyd and Van Acker 2003; Chauhan et al. 2006). However, both of the species we used are considered to have transient weed seedbanks and a relatively low persistence in the field, which may partially explain the high losses observed (Barralis et al. 1988; Goggin et al. 2012; Jensen 2009). High precipitation rates during this particular summer in all locations may also have prompted exceptionally high germination rates; this probably would not have occurred in a normal dry year. High moisture levels are also known to favor microbial activity and prompt seed decay (Wagner and Mitschunas 2008), which could also have contributed to the high seed losses reported here. Survival rates in the soil of other important weed species in cereal systems with more persistent weed seedbanks, such as corn poppy (Papaver rhoeas L.) or common lambs-quaters (Chenopodium album L.), should be expected to be higher than those reported for rigid ryegrass and catchweed bedstraw, and extending seed exposure to predators may be even more important to decrease those species’ seedbanks.

Overall, the results of this experiment showed that long-term seed loss due to predation was higher than loss caused by seed burial. This suggests that tilling the field immediately after crop harvest would have resulted in higher weed seed survival rates compared with leaving seeds on the soil surface exposed to predators. Both for transient and for more persistent weed seeds, leaving the fields untillled throughout the summer and maximizing seed exposure to predators seems to be the optimum weed management strategy during the fallow period in these systems. Decreasing the density of crop seeds on the soil surface by carefully adjusting the harvesting equipment has the potential to increase weed seed mortality to predation even further.

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