

## Research Article

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# Using a seed impact mill to limit waterhemp (*Amaranthus tuberculatus*) seed inputs in Iowa soybean

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## Abstract

Mounting cases of herbicide-resistant waterhemp [*Amaranthus tuberculatus* (Moq.) Sauer] in the U.S. Midwest have renewed the interest in nonchemical weed management strategies. Field experiments were conducted in 2021 and 2022 to quantify the effectiveness of a commercial combine equipped with a seed impact mill in preventing *A. tuberculatus* seed return to the soil seedbank in soybean [*Glycine max* (L.) Merr.]. *Amaranthus tuberculatus* seed shattering before crop harvest was quantified. *Amaranthus tuberculatus* started shattering seeds during the last week of August in both years. Overall, 51% of *A. tuberculatus* seeds were retained on the plant at harvest on October 23, 2021, compared with 61% at harvest on October 7, 2022. Viability of shattered *A. tuberculatus* seeds ranged from 84% to 94%. Additional seed shattering occurred when plants were disturbed by the combine header during soybean harvest, which caused 15% and 9% shattering in 2021 and 2022, respectively. *Amaranthus tuberculatus* seeds passed through the impact mill were grouped in three categories: no damage, moderate damage, and severe damage. In 2021, *A. tuberculatus* seeds with moderate damage had 26% lower germination and viability than seeds with no visible damage. In 2022, seed germination and viability of no-damage seeds did not differ from seeds with a moderate level of damage. No severely damaged seed germinated or tested viable in either year. Altogether, impact mill treatment reduced the number of germinable seeds by 87% compared with the no-impact mill treatment. These results indicate that seed impact mills can be a useful tool in Iowa soybean production to help manage multiple herbicide-resistant *A. tuberculatus* populations. However, *A. tuberculatus* seed shattering before crop harvest reduces the overall effectiveness of seed impact mills in preventing seedbank replenishments.

## Introduction

Waterhemp [*Amaranthus tuberculatus* (Moq.) Sauer] is one of the greatest weed problems in midwestern U.S. soybean [*Glycine max* (L.) Merr.] production (Van Wychen 2022). Herbicides have been a primary tool to control *A. tuberculatus*, resulting in widespread evolution of herbicide-resistant (HR) *A. tuberculatus* populations across the region (Heap 2024; Tranel 2021). More than 66% of *A. tuberculatus* populations from Iowa are resistant to inhibitors of acetolactate synthase, photosystem II, and enolpyruvylshikimate phosphate synthase (glyphosate) (Hamberg et al. 2023). This has substantially reduced herbicide options to control *A. tuberculatus* in soybean. Therefore, nonchemical weed management practices in conjunction with herbicides are needed to manage HR *A. tuberculatus* populations in this region.

Nonchemical weed control tactics such as tillage, cover crops, and reduced row spacing have proven effective in managing HR *A. tuberculatus* (Farmer et al. 2017; Yadav et al. 2023). However, the focus of these control tactics has been on preventing weed seedling establishment early in the growing season (Liebman and Gallandt 1997). Because late-season weed survivors/escapes rarely cause crop yield losses due to their inability to compete with previously established crop (Hartzler et al. 2004), they are often ignored on large commercial farms, despite their ability to produce large numbers of seeds (Bagavathiannan and Norsworthy 2012). Therefore, additional control tactics targeting seed inputs are needed to prevent seedbank replenishment.

Harvest weed seed control (HWSC) is a relatively new nonchemical weed control tactic that focuses on weed survivors/escapes. The HWSC method manages or destroys weed seeds at the time of crop harvest. One HWSC method is weed seed destruction using seed impact mills attached to the combine (Walsh et al. 2017). In this method, weed seed-bearing crop chaff is directed through high-impact mills that are integrated at the rear of combine. Several seed impact mills have been developed commercially, including Redekop™ Seed Control Unit, iHSD® Harrington Seed Destructor, Seed Terminator™, and WeedHOG™. These impact mills have been proven effective in damaging weed seeds retained on plants at the time of crop harvest (Schleich

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et al. 2023; Walsh et al. 2018). Seeds with visible damage are less likely to persist in the soil seedbank due to increased seed mortality (Davis et al. 2008; Gossen et al. 1998). Damage to the physical integrity of the seed may reduce seed germinability through two ways. First, it disrupts normal metabolic activity required for seed germination and survival (Gossen et al. 1998). Second, it reduces barriers for fungi and other microbial attacks (Gossen et al. 1998), which overwhelm the seed defense mechanisms and increase seed mortality (Davis et al. 2008). Little research has been conducted on the effectiveness of seed impact mills in managing troublesome weeds in U.S. production systems.

A high percentage of weed seed retention on the plant at the time of crop harvest is essential for seed impact mills to be a viable option in reducing weed seed inputs into the soil seedbank. Weed seed shattering (natural shedding of seeds when they ripen) before crop maturity reduces the proportion of seeds captured by the combine at crop harvest, hence lowering the effectiveness of HWSC methods. Seeds that are retained on the mother plant at the time of crop maturity may not enter the combine due to seed shattering during the harvesting process. When a combine header touches the plant, the mechanical disturbance created by the combine header can increase weed seed shattering (Winans et al. 2023; personal observations). Data on the percentage of *A. tuberculatus* seeds shattered before crop harvest and during harvest are lacking. The objectives of our study were (1) to quantify *A. tuberculatus* seed shatter timing and seed viability before crop harvest; (2) to quantify *A. tuberculatus* seed shattering caused by the mechanical disturbance of combine header during harvest; and (3) to evaluate the effects of a seed impact mill on the visible *A. tuberculatus* seed damage, germination, and viability.

## Materials and Methods

### Experimental Site

Field experiments were conducted in 2021 and 2022 on a commercial farm near Gilbert, IA (42.113298°N, 93.609298°W). Fields used in the experiments had been under corn (*Zea mays* L.)–soybean rotation for at least 10 yr and had a history of high levels of *A. tuberculatus*. Before the experiments, the fields were chisel plowed in the fall, and a field cultivator was used the following spring to prepare the seedbed.

Each year, soybean resistant to 2,4-D, glufosinate, and glyphosate was planted in 76-cm-wide rows at 370,660 seeds ha<sup>-1</sup>. In 2021, soybean ('Hoegemeyer 2660 E', Hoegemeyer®, Hooper, NE 68031) was planted on May 8. In 2022, soybean ('P22T18E', Pioneer®, Johnston, IA 50131) was planted on May 22. Each year, a preemergence herbicide program consisting of S-metolachlor (1.5 kg ai ha<sup>-1</sup>) + sulfentrazone (0.2 kg ai ha<sup>-1</sup>) was applied on the day of planting. No postemergence herbicide was applied. Soybean was harvested on October 23 in 2021 and October 7 in 2022 using a John Deere S680 combine (Moline, IL 61265). The combine was equipped with a seed impact mill (Redekop™ Seed Control Unit, Redekop Manufacturing, Saskatoon, SK S7K 3J7, Canada) (Figure 1).

An experimental area measuring 107 m by 91 m was selected in the soybean field uniformly infested with *A. tuberculatus*. The experimental area was divided into 10 plots arranged in a completely randomized design. Each plot was 10.7-m wide (equivalent to the width of the commercial combine header) and 91-m long. Records of average air temperatures and total precipitation during 2021 and 2022 growing seasons are summarized in Table 1.

**Table 1.** Average air temperature and total precipitation during 2021 and 2022 growing seasons on a commercial farm in Gilbert, IA.<sup>a</sup>

	Average temperature			Precipitation		
	2021	2022	30-yr avg.	2021	2022	30-yr avg.
	C			mm		
April	11	7	10	12	111	102
May	16	17	16	64	109	132
June	24	23	22	43	179	132
July	24	24	23	55	107	97
August	23	23	23	100	121	119
September	20	18	18	91	43	79
October	13	11	11	120	150	74
Total	—	—	—	485	820	735

<sup>a</sup>Temperature and precipitation data were obtained online from the Iowa State University Iowa Environmental Mesonet website: <https://mesonet.agron.iastate.edu/agweather>.

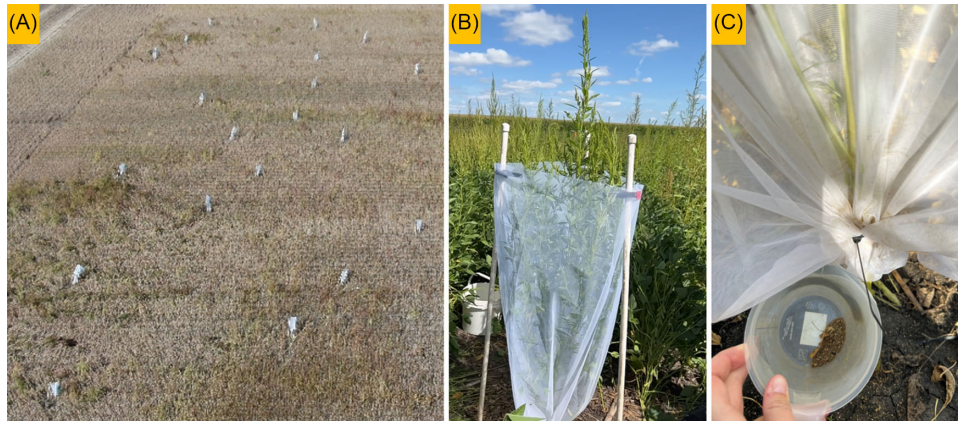


**Figure 1.** The Redekop™ Seed Control Unit/seed impact mill (a harvest weed seed control method) installed on a John Deere S680 combine in 2021 on a commercial farm in Gilbert, IA. The figure shows the rear of the combine without seed impact mill (A); impact mill installed to the combine (B); weed seed-bearing chaff exiting through the impact mill (C).

### Experimental Methods and Data Collection

#### Preharvest Measurements

*Amaranthus tuberculatus* density and seed production were recorded the day before soybean harvest to quantify the *A. tuberculatus* infestation levels. *Amaranthus tuberculatus* density was measured by counting seed-producing *A. tuberculatus* plants from 10 randomly placed 1-m<sup>2</sup> quadrats in each plot. *Amaranthus tuberculatus* seed production was measured by carefully harvesting 4 plants at random in each plot and drying them in an air-dryer at 25 C for 2 wk. Plants were then hand threshed and cleaned with handheld sieves. An air-column blower (Seedburow® Equipment, Des Plaines, IL 60018) was used to further clean seeds from fine plant debris. Four subsamples of 0.1 g of seed were counted to determine the average seed weight. Then, seeds per sample were calculated by dividing the total sample weight by the average seed weight.



**Figure 2.** Procedure used to estimate *Amaranthus tuberculatus* seed shattering over time in 2021 and 2022 on a commercial farm in Gilbert, IA. (A and B) Female *A. tuberculatus* plants encased in custom-designed bags in a soybean field. (C) Shattered *A. tuberculatus* seeds being collected in a plastic container at a weekly interval.

### Seed Shatter Experiment

Two female *A. tuberculatus* plants representative of each plot (based on visual assessments) were selected to quantify natural seed shatter before harvest. Plants were individually encased in seed traps at the seed development stage. The seed traps were custom designed by making an open-ended bag from Noseum Mosquito Netting Fabric (Online Fabric Store, West Springfield, MA 01089). The traps were then placed around the plant with the bottom end closed around the plant's stem using a plastic tie. The other end of the bag was kept open and secured around the plant using three PVC pipes driven into the soil. The trap design allowed free air movement through the plant canopy (Figure 2).

*Amaranthus tuberculatus* seed shattering was recorded on a weekly basis for all plants. *Amaranthus tuberculatus* seed collection started on August 28, 2021, and September 2, 2022, and ended on October 20, 2021, and October 7, 2022. Shattered seeds were collected by opening the bottom end of the bag and collecting seeds in a plastic container (Figure 2C). At soybean harvest, *A. tuberculatus* plants were cut at the ground level and dried in an air-dryer at 25 C for 2 wk. The samples were cleaned, and seeds were counted using the method described earlier.

To quantify the viability of *A. tuberculatus* seeds shattered over time, a germination test using 50 seeds from each observation time was conducted. Seeds were soaked in distilled water and stored at 4 C (wet-chilling) for 2 wk to break seed dormancy (Leon and Owen 2003) and then air-dried at room temperature (25 C) for 2 d. Dry seeds were put between two filter papers in 9-cm-diameter petri dishes and moistened with 7 ml of distilled water. Petri dishes were placed in a growth chamber (Percival GR36LC8, Perry, IA 50220) set at 32 C day and 22 C night temperatures with a day and night cycle of 14 and 10 h, respectively. Seed germination was observed for 4 wk. Germinated seeds were counted and removed from petri dishes at 1-wk intervals. At the end of observation period, nongerminated seeds (potentially viable) were tested for viability using the imbibed seed crush test (Borza et al. 2007). Seeds that collapsed under gentle pressure from forceps were considered as nonviable, whereas firm seeds were considered as viable. The proportion of viable seed was calculated by adding the number of seeds germinated plus the number of seeds rated as viable in the crush test divided by the total number of seeds evaluated.

### Header Loss Experiment

Ten female *A. tuberculatus* plants in each plot were selected to measure seed shattering due to combine header during crop

harvest. Two plastic pans (105 cm by 70 cm) were placed underneath each plant to capture the shattered seeds. Pans were kept underneath until the plant was cut and fed in the conveyer and the combine header completely passed over the pans. Once this process was completed, the combine was stopped and backed up and pans were safely removed. Shattered seed samples were transferred to paper bags. Because the sampled plant was destroyed in this collection method, the initial number of seeds present on the original plant being harvested by the combine header could not be counted. A second plant similar in height and canopy diameter to the original plant was selected and cut to estimate the initial number of seeds present on the plant used to measure header loss. The samples were cleaned, and seeds were counted using the method described earlier. The number of seeds entering the combine were calculated by subtracting the number of seeds shattered due to combine header from the total number of seeds present on the comparable plant at the time of harvest.

### Seed Impact Mill Experiment

Eight plots were grouped in four blocks each consisting of two plots to quantify the effectiveness of the seed impact mill. The impact mill was engaged and disengaged from the combine during harvest to create two treatments, impact mill versus no impact mill. Treatments were assigned randomly in each block. Threshed residue from the rear of the combine was collected in plastic trays (70 cm by 105 cm) during soybean harvest (Figure 3). Trays were placed on the ground in a zigzag pattern once the combine header had passed, but before the threshed residue was returned to the field. Eight trays were used in each plot. Threshed residue was placed in paper bags for further processing. Samples collected from the no-impact mill treatments were cleaned by using the method described for the seed production data. Because samples from impact mill treatments contained finely ground chaff-seed mixture, a different method was used to separate *A. tuberculatus* seeds from the chaff without blowing away the broken seed pieces. Samples were initially hand sieved to separate large plant debris from chaff-seed mixture. Then samples were placed on an experimental vibratory separator (Gregg and Billups 2010) that separated intact and broken seeds from fine chaff. Seeds were inspected under a microscope to assess visible damage and were grouped in three categories: no damage (<10% damage), moderate damage (10% to 30%), and severe damage (>30%). Seeds with no



**Figure 3.** Sample-collection procedure to estimate the seed impact mill damage effectiveness on *Amaranthus tuberculatus* seeds during soybean harvest in 2021 and 2022 on a commercial farm in Gilbert, IA. (A) Plastic trays thrown to capture weed seed-bearing soybean chaff exiting the impact mill. Once the combine completed the pass (B), the collected material was transferred to the paper bags for further processing (C).

visible damage or only surface abrasions were included in the no-damage category.

The seed viability test method described earlier was used to determine seed germination and viability for all seeds collected in the no-impact mill or impact mill treatments. Seeds in the no-damage category plus seeds that tested viable in the moderate-damage category were considered germinable and used to calculate the damage effectiveness of seed impact mill (Equation 1).

$$E = \frac{A - P}{A} \times 100 \quad [1]$$

where  $E$  is the percent damage effectiveness of seed impact mill, and  $A$  and  $P$  are the number of germinable seeds in no-impact mill and impact mill treatments, respectively.

### Data Analysis

Data on *A. tuberculatus* density, seed production, header loss, and seed impact mill effectiveness were compared using a two-sample  $t$ -test ( $\alpha = 0.05$ ) in SAS v. 9.4 software (SAS Institute, Cary, NC 27513). All the seed shatter, seed germination, and viability data were analyzed using PROC GLM in SAS v. 9.4 software.

Cumulative *A. tuberculatus* seed retention was analyzed in the statistical programming language R (R Core Team 2019) using the R extension package DRC (Ritz et al. 2019). A three-parameter log-logistic model was fit using Equation 2 (Knezevic et al. 2007) to plot the percent *A. tuberculatus* seed retention over time:

$$y = \frac{d}{1 + \exp\{b[\log x - \log e]\}} \quad [2]$$

where  $y$  denotes the percentage of seed retained on the mother plant (relative to the start of observation period) and  $x$  denotes the time (week). Parameter  $d$  denotes the upper limit. Parameter  $e$  denotes the  $t_{50}$  (time required to reduce percentage of seeds retained on the plant by 50%). Parameter  $b$  denotes the relative slope around  $e$ . Additionally, the value of  $t_{10}$  was calculated using the ED function of the DRC package.

### Results and Discussion

Because the experimental years differed in soybean planting, harvesting, and seed-shattering collection dates, data for each response variable were analyzed by year. Spring of 2022 was wetter and colder than the spring of 2021, which delayed soybean planting by 2 wk. The average air temperature for the 2021 and 2022

growing seasons ranged from 11 to 24 C and 7 to 24 C, respectively (Table 1). Total precipitation during the 2021 growing season (485 mm) was lower than that of 2022 growing season (820 mm). Average air temperature in October (typical soybean harvest period) 2021 and 2022 was 13 and 11 C, respectively. Total precipitation during October 2021 and 2022 was 120 and 150 mm, respectively.

In both years, female *A. tuberculatus* density and seed production was uniform across the seed impact mill treatments. *Amaranthus tuberculatus* density was lower in 2021 (less than 1 plant  $m^{-2}$ ) than in 2022 (8 plants  $m^{-2}$ ). Similarly, *A. tuberculatus* seed production was lower in 2021 (17,300 to 29,200 seeds  $m^{-2}$ ) than in 2022 (1 million to 1.1 million seeds  $m^{-2}$ ). Hartzler et al. (2004) previously reported *A. tuberculatus* produced more than 1 million seeds  $plant^{-1}$  in Iowa soybean. The high *A. tuberculatus* density and seed production in 2022 was likely due to high precipitation during the growing season, specifically in June and July (Table 1), which is the peak *A. tuberculatus* emergence period (Hartzler et al. 1999).

### Seed Shatter

*Amaranthus tuberculatus* started shattering seeds between August 21 and 28 in 2021, and August 26 and September 2 in 2022 (Table 2). During the first week of observation, *A. tuberculatus* shattered 870 seeds  $plant^{-1}$  in 2021 compared with 310 seeds  $plant^{-1}$  in 2022. The highest level of *A. tuberculatus* seed shattering in 2021 (29,570 seeds  $plant^{-1}$ ) occurred between October 8 and 15 compared with September 21 and 28 in 2022 (43,150 seeds  $plant^{-1}$ ). The number of *A. tuberculatus* seed shattered between each collection date did not increase or decrease consistently over time. The variation in the number of seeds shattered between the collection dates could be due to occurrence of brief weather events such as windstorms, temperature fluctuations, or rainfall events (Forcella et al. 1996; Nielsen and Vigil 2017).

The percentage of *A. tuberculatus* seeds retained on the plant decreased over time in both years (Figure 4). Overall, 51% of *A. tuberculatus* seeds were retained on the plant at the time of soybean harvest in 2021, which occurred on October 23, compared with 61% at 2022 harvest, which occurred on October 7 (Figure 4). *Amaranthus tuberculatus* plants retained >90% of total seeds until 3 wk after the initial seed shattering started in each year (Table 3). Fifty percent of *A. tuberculatus* seed shattering occurred 8 wk after the initial seed shattering in 2022 compared with 7 wk in 2021. Bennett et al. (2023) previously reported that 90% of *A. tuberculatus* seeds were retained on the plant until September 19 or 2 wk before soybean harvest. However, seed retention declined to 70% at soybean harvest.

*Amaranthus tuberculatus* seed viability for all shattering timings ranged from 84% to 94% in both the years. The high

**Table 2.** *Amaranthus tuberculatus* seed shatter at each observation date and shattered seed viability in soybean in 2021 and 2022 on a commercial farm in Gilbert, IA.<sup>a</sup>

Date	2021		Date	2022	
	Seeds shattered	Seed viability		Seeds shattered	Seed viability
	no. plant <sup>-1</sup>	%		no. plant <sup>-1</sup>	%
August 28	870 f	85 bc	September 2	310 d	90 a
September 5	1,830 ef	94 a	September 8	5,010 cd	90 a
September 14	7,840 de	89 ab	September 15	6,680 c	94 a
September 20	22,260 b	93 a	September 21	16,500 b	91 a
September 27	13,300 cd	84 c	September 28	43,150 a	89 a
October 8	15,540 c	91 a	October 7	22,690 b	91 a
October 15	29,570 a	88 abc	—	—	—
October 20	6,520 ef	92 a	—	—	—

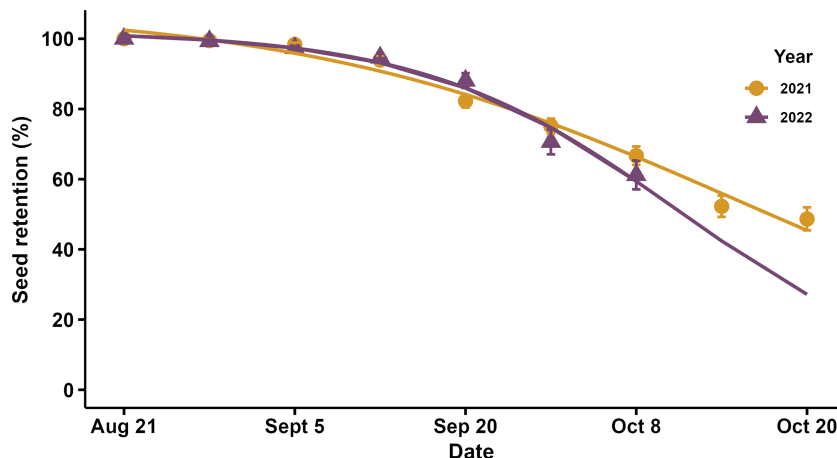
<sup>a</sup>Treatment means within a column with the same letter are not significantly different (LSD test,  $\alpha = 0.05$ ).

**Table 3.** Estimated parameter values using the log-logistic model (Equation 2) to quantify the percentage of *Amaranthus tuberculatus* seeds retained on the plant over time in soybean in 2021 and 2022 on a commercial farm in Gilbert, IA.

Experiment year	Parameter estimates ( $\pm$ SE) <sup>a</sup>			
	<i>b</i>	<i>t</i> <sub>10</sub>	<i>t</i> <sub>50</sub>	<i>d</i>
2021	2.58 (0.21)	3.23 (0.21)	7.58 (0.17)	100.60 (1.40)
2022	3.50 (0.44)	3.60 (0.21)	6.74 (0.22)	99.68 (1.31)

Values in parentheses represent standard errors of the means.

<sup>a</sup>Parameter *b* is the relative slope around *t*<sub>50</sub>. Parameter *t*<sub>50</sub> is the time (in weeks) required to reduce the percentage of seeds retained on the plant by 50%. Similarly, *t*<sub>10</sub> is the time (in weeks) required to reduce the percentage of seeds retained on the plant by 10%. Parameter *d* is the maximum seed retention (%) at start of the observation period.



**Figure 4.** Percentage of *Amaranthus tuberculatus* seeds retained on the plant over time in soybean in 2021 and 2022 on a commercial farm in Gilbert, IA. Curves were generated using a three-parameter log-logistic model (Equation 2). Symbols on the curves are the observed means of the replicates.

levels of seed viability even in early-shattered seeds could be explained by the fact that *A. tuberculatus* seeds can become viable 7 to 9 d after pollination (Bell and Tranel 2010). However, the exact series of events that led to early shattering of viable *A. tuberculatus* seeds needs to be investigated. These results indicate that early shattered seeds contribute to soil seedbank replenishment even in the presence of HWSC methods.

### Header Loss

In addition to *A. tuberculatus* natural seed shattering, seeds were also shattered by the mechanical disturbance created by the combine header during soybean harvest. During this process,

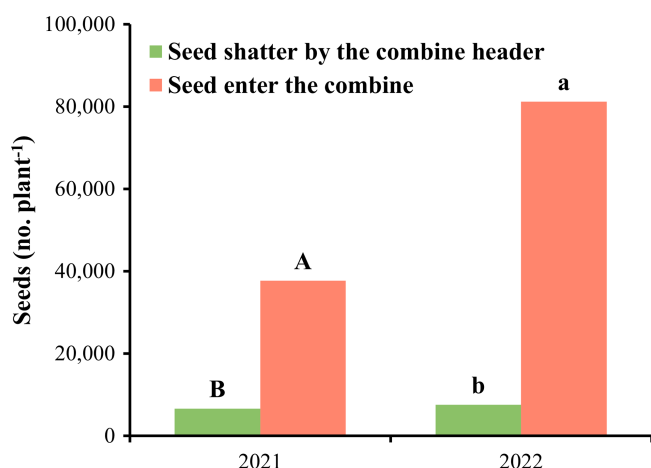
*A. tuberculatus* shattered 15% and 9% of the seeds that were retained on plants in 2021 and 2022, respectively (Figure 5). Winans et al. (2023) reported 22% to 40% *A. tuberculatus* seed shatter when plants were disturbed by the combine header during soybean harvest. Schleich et al. (2023) reported that *A. tuberculatus* seed shattering due to the combine header averaged <3%, which might have been influenced by the low level of *A. tuberculatus* seed retention (30%) at the time of soybean harvest. Factors such as plant physiological characteristics and growth stage at harvest, plant interaction with insects and pathogens, weather events, and combine disturbance can affect weed seed shattering (Abul-Fatih et al. 1979; Goplen et al. 2016; Hobson and Bruce 2002; Shirtliffe et al. 2000).

**Table 4.** *Amaranthus tuberculatus* seed visible damage, germination, and viability of the seeds collected from the threshed residue during soybean harvest in 2021 and 2022 on a commercial farm in Gilbert, IA<sup>a</sup>.

	Levels of visible damage <sup>b</sup>	Seeds in visible damage category		Seed germination		Seed viability	
		2021	2022	2021	2022	2021	2022
No impact mill	No damage	—	—	50 a	23 a	63 a	36 a
Impact mill	No damage	18 b	4 a	49 a	17 ab	56 a	22 ab
	Moderate damage	27 ab	5 a	23 b	12 bc	30 b	13 bc
	Severe damage	55 a	91 b	0 c	0 c	0 c	0 c

<sup>a</sup>Treatment means within a column with the same letter are not significantly different (LSD test,  $\alpha = 0.05$ ).

<sup>b</sup>Seeds collected from the threshed residue after passing through the seed impact mill were grouped in three categories based on the levels of visible damage of the seed: no damage = <10% visible damage; moderate damage = 10% to 30% damage; severe damage = >30% damage.



**Figure 5.** *Amaranthus tuberculatus* seeds shatter when shaken by the combine header (John Deere S680) during soybean harvest in 2021 and 2022 on a commercial farm in Gilbert, IA. Bars within a pair with different letters are significantly different (two sample *t*-test,  $\alpha = 0.05$ ).

### Seed Impact Mill

The impact mill caused different levels of damage (*E*) to *A. tuberculatus* seed (Table 4). In 2021, 82% of *A. tuberculatus* seeds had >10% visible damage compared with 96% in 2022. Schwartz-Lazaro et al. (2017) have previously reported >95% damage of *A. tuberculatus* seeds when crop chaff and seeds passed through stationary impact mills. The germinability and viability of intact seeds in impact mill treatments did not differ from intact seeds collected in no-impact mill treatments (Table 4). However, it is possible that those seeds may have not entered the impact mills but passed through the straw-chopper instead. Intact seed germination and viability ranged from 17% to 50% and 22% to 63%, respectively.

Visible damage caused by the impact mill reduced *A. tuberculatus* seed germinability and viability percentages (Table 4). In 2021, *A. tuberculatus* seeds with a moderate level of visible damage had 26% lower germination and viability than seeds with no visible damage. In 2022, seed germination and viability of intact seeds and seeds with a moderate level of damage did not differ. No seed in the severe damage category germinated or tested viable in either year. Hauhouot-O'Hara et al. (1998) reported that an increasing level of physical damage to weed seeds greatly reduces their germinability and viability. In 2021, the impact mill treatment resulted in 83% less germinable seed

compared with the no-impact mill treatment. In 2021, the number of germinable seeds in the impact mill treatment (120 seeds  $m^{-2}$ ) was 83% lower than in no-impact mill treatment (720 seeds  $m^{-2}$ ). Similarly, in 2022, the number of germinable seeds in the impact mill treatment (2,420 seeds  $m^{-2}$ ) was 90% lower than in the no-impact mill treatment (23,520 seeds  $m^{-2}$ ).

### Management Implications

These results indicate that a seed impact mill is highly effective in damaging *A. tuberculatus* seeds that enter the combine, hence reducing the return of germinable seeds into the soil seedbank. Although the impact mill did not severely damage all of the *A. tuberculatus* seeds, moderate damage to seeds was effective in reducing seed germination and viability in controlled conditions. Furthermore, seeds with moderate damage are less likely to persist in soil seedbank due to increased seed mortality (Davis et al. 2008; Gossen et al. 1998).

Weed survivors are becoming more common in production fields due to the widespread occurrence of multiple herbicide-resistant populations (Bagavathiannan and Norsworthy 2012). Maintaining a low weed seedbank density is critical for herbicide-resistance management (Neve et al. 2011). Mainstream weed management programs for U.S. soybean production do not include a late-season weed control strategy. As a result, weed escapes/survivors are the primary source of seedbank replenishment. Implementation of seed impact mills in the current system would diversify the weed control strategies in use and might delay the development of HR populations. For example, Somerville et al. (2018) estimated that reductions in weed seed inputs by seed impact mills can delay the development of HR populations by 5 to 8 yr. Therefore, implementation of a seed impact mill in the Iowa soybean production system can be an effective strategy for the management of multiple herbicide-resistant *A. tuberculatus* populations.

Despite high effectiveness of seed impact mills in reducing the number of germinable seeds, seeds shattering before entering the combine reduce overall effectiveness of seed impact mills in preventing seedbank replenishments. These losses mainly occur through natural seed shatter and seed shatter due to the combine header. Seeds that enter the combine are also subjected to losses. It is possible that weed seeds may bypass the impact mills, instead escaping through the straw-chopper and/or being carried to the grain tank.

High levels of seed viability in seeds shattering before crop harvest emphasizes that additional adjustments to the crop harvest

practice would be required to maximize the proportion of weed seeds entering the combine. One of the biggest factors likely to influence the percentage of weed seed entering the combine is the time of crop harvest. Harvesting soybean at earlier dates would reduce the proportion of *A. tuberculatus* seeds that naturally shatter. This can be achieved by prioritizing harvest-ready fields with the highest levels of *A. tuberculatus* infestation during the harvesting season. The combines should be cleaned to reduce weed seed movement between fields. Furthermore, weed seed shattering due to the combine header can be minimized by modifying the combine header. In the past, efforts have been made in combine header designs to reduce crop seed shattering during the crop harvest (Henry et al. 2008; Hobson and Bruce 2002; McKay et al. 2003). Similar efforts may have the potential to reduce mechanical shattering of weed seeds associated with the combine header during crop harvest.

Implementation of HWSC methods in Iowa cropping systems is not a replacement of existing weed control tactics but rather an expansion of the weed management toolbox. All weed control tactics have limitations, and overreliance on a single tactic may increase weed control failures. It is likely that overreliance on HWSC methods will lead to the selection of early seed shattering weed biotypes (Somerville and Ashworth 2024). Other nonchemical weed control tactics such as cereal rye (*Secale cereale* L.) cover crop and narrow-row soybean have proven effective in managing HR *A. tuberculatus* in soybean, and therefore should be used in conjunction with HWSC methods to spread the risk of weed control failures (Liebman and Gallandt 1997; Yadav et al. 2023). Future research should focus on the long-term impact of integrating HWSC methods on *A. tuberculatus* life-history traits including its seedbank persistence.

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