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Article

Weed Seed Bank Dynamics: Weed Seed Bank Modulation Through Tillage and Weed Management

Dinâmica do Banco de Sementes de Plantas Daninhas: Modulação do Banco de Sementes de Plantas Daninhas por meio do Preparo do Solo e Manejo de Plantas Daninhas

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ABSTRACT - A field trial was conducted with the aim of assessing weed seed bank status of soil under maize cultivation. It was worked out by sampling from soil corer at different depths ranging from 0-5, 5-10, 10-15, 15-20 and 20-25 cm. Weed seed bank was monitored with a cultivator, a rotavator, a mouldboard plough (MB) and a chisel plough in tilled plots under Atrazine 330 EC at 1.00 kg a.i. ha^{-1} at 20 DAS, hand weeding at 20 and 40 DAS and no weeding. Sixteen weed species were identified in soil samples, four species contributing about 72% of the total weed seed bank. Tillage system was more important determinant of weed seed density than the weed management practices. Movement pattern of weed seeds by all tillage treatments differ significantly over three weeding management practices at 0-5 cm soil core. Horse purslane (28%) and jungle rice (20%) were with high relative abundance. In both years, change in weed seed bank in upper 0-5 cm soil was significant as seeds germinating from this layer were either removed by hand or killed with chemical or left unchecked. Only chisel plough tapped the soil core below 20 cm and caused a small reduction in weed seed density. Mouldbold plough effectively buried weed seeds below 10 cm but not beyond 20 cm and hand hoeing reduced weed seed bank by killing weeds at seedling stage.

Keywords: atrazine spray, hand weeding, tillage, vertical movement.

RESUMO - Um experimento de campo foi conduzido com o objetivo de avaliar o status de um banco de sementes de plantas daninhas de solo sob cultivo de milho. Ele foi realizado por amostragem de solo com um trado em diferentes profundidades, que variaram entre 0 5, 5-10, 10-15, 15-20 e 20-25 cm. O banco de sementes de plantas daninhas foi monitorado com o uso de cultivador, enxada rotativa, arado de aiveca (AA) e arado escarificador em parcelas lavradas sob Atrazina 330 EC a 1,00 kg i.a. ha^{-1} aos 20 DAS, com capina manual aos 20 e 40 DAS e com ausência de capina. Foram identificadas 16 espécies de plantas daninhas nas amostras de solo, das quais quatro contribuíram com cerca de 72% do total do banco de sementes. O sistema de preparo foi fator mais importante na determinação da densidade de sementes de plantas daninhas do que as práticas de manejo. O padrão de movimento das sementes de plantas daninhas, em todos os tratamentos de preparo de solo, difere significativamente em relação às três práticas de manejo dessas plantas na amostra de solo na camada de 0-5 cm. Bredo (28%) e capituva (20%) apresentaram alta abundância relativa. Em ambos os anos, houve mudança significativa no banco de sementes de plantas daninhas na camada superficial do solo (0 5 cm), uma vez que as sementes germinadas a partir desta camada foram

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removidas manualmente ou mortas com produtos químicos ou não controladas. Somente o arado escarificador atingiu o núcleo do solo abaixo de 20 cm e causou pequena redução na densidade das sementes da planta daninha. O arado de aiveca enterrou efetivamente as sementes de plantas daninhas abaixo dos 10 cm, porém não além dos 20 cm, enquanto a capina manual reduziu o banco de sementes dessas plantas, eliminando-as na fase de plântulas.

Palavras-chave: spray de atrazina, capina manual, sistema de preparo, movimento vertical.

INTRODUCTION

Weeds are one of the major causes of poor yield in different cropping patterns of Pakistan (Ali et al., 2011). Soil weed seed bank or seed pool is a critical factor that determines above-ground floristic composition and weed density in agricultural landscapes (Norris, 2007). The quantitative as well as qualitative measures of weed seed bank raise awareness of the physical history of past successful or failed attempts of cropping systems. They also help growers to predict the extent to which they are going to face weed problems and how they will drastically affect crop yield and quality in the future (Cardina et al., 2002). Along with tillage, other practices such as mulching, crop rotation and herbicide application also affect the fate of weeds in the upcoming crop (Swanton et al., 2008; Nichols et al., 2015).

Tillage induces changes in soil microclimate, e.g., in moisture, temperature and light (Prior et al., 2004; Alvaro-Fuentes et al., 2007). Seeds respond highly to these vertical gradients. Seed position in the soil is one of the major factors influencing seed germination and seedling emergence (Kevin et al., 2015) which is disturbed by tillage. Jun et al. (2002) negatively related seed germination and burial depth of ten *Calligonum* species and argued that the deeper the seeds in sand, the lower and slower their germination and seedling emergence will be.

Herbicide use can alter the spectrum and size of a weed seed bank in the soil. Herbicides have remained the most effective weed management tool of the 20th century. They are very effective at reducing weed density and soil seed bank (Hossain et al., 2014). Weed seed bank densities tend to be greater in organic cropping systems than in systems reliant on herbicides. In production systems which use herbicides as the main tool for weed control, seed bank densities range typically between 1000 and 4000 seeds m^{-2} (Blackshaw et al., 2004). High herbicide input decreases soil seed bank buildup (Ranjit et al., 2007). Manual weed control is the oldest approach that has changed progressively from hand pulling to use of a wide range of implements e.g. harrows, cultivators and mowers (Liebman et al., 2001). The most effective weed control comes from crushing, breaking, cutting and crimping the stems and roots of weeds, which can be achieved by hand weeding (Creamer and Dabney, 2002).

Managing weed seed bank to lessen the number of viable weed seeds in the soil is one of the most important control strategies (Gallandt, 2006). The present study describes density, distribution and vertical movement of weed seeds under different tillage systems and weed management practices and their effect on future soil weed seed bank status. The findings of this study could be applied to predict floristic composition of weed species from the soil seed bank, and they can be manipulated to alter weed seed distribution by different agronomic approaches.

MATERIALS AND METHODS

Study site and design: A two-year experiment was carried out at the Agronomic Research Area, University of Agriculture, Faisalabad. The experiment was laid out in completely randomized block design with split arrangement and three replications, and net plot size of 4.5 m x 6 m. Soil of the field in both years was well drained loamy soil with 1% organic matter, and soil pH was 8.5 (slightly alkaline). The field had been under spring and autumn maize (*Zea mays* L.) crop for the last five years. Four tillage levels were assigned to the main plots; (T_1) cultivator alone twice, (T_2)

rotavator once + cultivator twice, (T_3) mouldboard plough once + cultivator twice and (T_4) chisel plough once + cultivator twice. Weed management was placed in the sub-plots with three levels; (W_1) herbicide spray (Atrazine 330 EC at 1.00 kg a.i. ha^{-1}) at 20 DAS, (W_2) hand weeding with shovel at 20 and 40 DAS and (W_3) weedy check (no weeding).

Soil Sample Collection: Soil samples were taken from 0-5, 5-10, 10-15, 15-20 and 20-25 cm soil depth with a soil auger (corer) with 76 mm internal diameter before sowing the crop in the 1st week of August. Samples were taken from five different places within the plot (four at corners, leaving 1 meter space at the edge. and one from the middle). Craft paper bags with normal size were used to carry the soil samples to the laboratory. Each sample taken from a particular plot was thoroughly mixed and stored in cooled chambers at 0 °C to halt any metabolic activity. Maize hybrid (Monsanto-DK 919) was manually sown in the first week of August on the ridges using a seed rate of 25 kg ha^{-1}. Row to row distance was maintained at 75 cm and plant to plant distance, at 25 cm. Thinning was performed to ensure plant to plant Nitrogen was applied at the rate of 250 kg N ha^{-1}. The phosphorus and potassium fertilizer were applied at the rate of 120 and 125 kg ha^{-1}, respectively. The field was irrigated ten days after germination. All (8) irrigations were applied through flooding. Soil samples were again collected the day after harvesting to avoid any further addition or withdrawal to the weed seed bank, and the same procedure was followed after the crop harvest (December 2013-2014), and the samples were properly tagged.

Seed Sorting and Data Collection: Soil samples were collected before sowing and after harvesting of maize. Seeds from the samples were sorted in two phases; In the first phase i.e. germination, each soil sample was placed separately in 3 cm deep containers (30 cm x 15 cm) and watered daily (Mesquita et al., 2015) by a sprinkler in the laboratory. Seedling emergence was monitored over a period of 4 months (4 cycles). Soil was stirred once a month after cessation of further weed seedling emergence (Jha et al., 2014). Seedlings were identified by species, counted and removed after recording. After one week, trays were re-watered to allow further germination. They aim was to have maximum germination so that the number of viable seeds of each and every species could be counted as they germinated.

In the second phase i.e. elutriation, the soil samples from the germination trays were washed with water and the seeds were collected in a 500 µm (# 35) sieve through pouring water. After the mud was removed by water flowing through the sieve, seed samples were shifted from the sieve to fine mesh cloth and were tied with twist ties. The samples were put into an oven at 40 °C overnight to dry. Seeds were sorted at first by a magnifying glass and then under microscope, manually (Fogliatti, 2003; Forcella et al., 2004). Seeds were sorted on the basis of species, counted and put into small glass jars after the numbers were recorded.

Data Analysis: The number of seeds found in each sample in both phases (germination and elutriation) was summed and recorded in data columns for each treatment to calculate weed seed density (seeds m^{-2}). Significance of different treatments was checked by LSD test at $p \leq 0.05$ (Steel et al., 1997) using Statistix 10.0.

RESULTS AND DISCUSSION

Relative abundance (%)

Form the soil samples taken, seeds of 16 different weed species were recovered (Figure 1) with variable relative abundance (No. of seeds in a species/Total no. of seeds x 100). Out of the sixteen weed species found, horse purslane (*Trianthema portulacastrum*) had the maximum relative abundance (27%) followed by weed seeds of jungle rice (*Echinochloa colona)* with relative abundance of 20% of the entire weed seeds recorded. Weed seeds of false amaranth (*Digera muricata*) and johnson grass (*Sorghum halepense*) were found to have relative abundance of 16% and 14%, respectively. The remaining twelve different types of weed species were recorded as the least abundant, with relative abundance ≤ 4%. Among minor weeds, seeds of bermuda grass (*Cynodon dactylon*), barnyard grass (*Echinochloa crus-galli*), goose grass (*Eleusine indica*), false amaranths (*Digera muricata*), lambsquarters (*Chenopodium album*) and broadleaf dock (*Rumex obtusifolius*) were found to account for only 1% of presence in the soil weed seed bank.

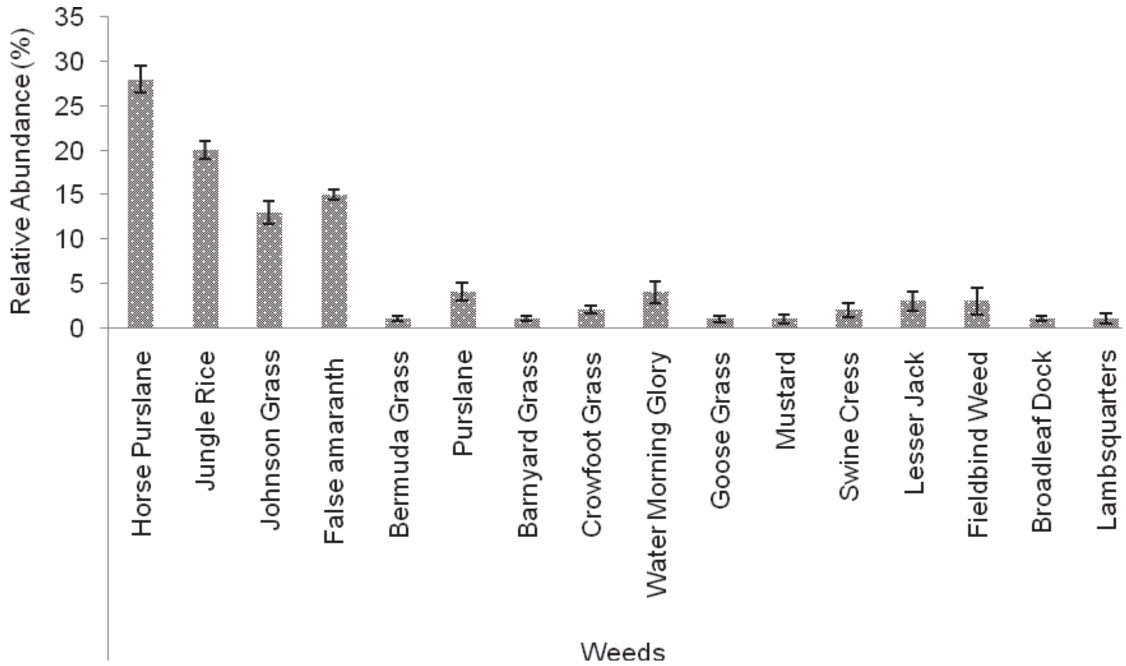

Figure 1 - Relative abundance of weed species found in both years (2013-14).

Species composition

Sixteen (16) weed species were found in the experimental plots either in weed flora or in the soil weed seed bank or in both (weed and soil seed bank). Out of those sixteen weed species, ten were found in weed flora and in the weed soil seed bank in different plots while only six weed species (water morning glory, false amaranth, lesser jack, field bind weed, broadleaf dock and lambsquarter) were found to be present only in the soil weed seed bank (Table 1). The experimental plots treated with chisel plough and MB plough were found to have maximum weed seed diversity at deeper soil cores (15-20 and 20-25 cm). It was not surprising that all the species did not appear in all soil layers. Weed species found in the MB plough system were found in all tillage systems.

Tillage-by-weeding interaction

There was a significant interaction between different tillage systems and weed management practices for the weed seed bank of all the species (16) at surface (0 5 cm) throughout 2013 (Table 2). The highest weed seed density ($p \leq 0.05$) was recorded in cultivator x no weeding, and chisel plough x no weeding (13,852 and 13,019 seeds m^{-2}, respectively). In MB plough tilled experimental plots which were hand-weeded at 20 and 40 DAS, total weed seed density was significantly lower (5,187 seed m^{-2}). At soil depth beyond 0-5 cm, none of the weed management treatments was effective in altering weed seed density. However, all the four tillage systems modified weed seed position depending on their effective depth and action. In MB plough, for example, least weed seed density was recorded (7,304 seeds m^{-2}) at the 5-10 cm depth. Contrarily, at the 10-15 cm depth, the lowest weed seed density (5,404 seeds m^{-2}) was recorded in case of cultivator only, as shallow tillage by cultivator did not stir the soil and seed density might be lower because of the soil was settled. Soil core of 15-20 cm with MB plough as tillage system showed maximum weed seed density (5,976 seed m^{-2}) while other three tillage systems were at par with each other with the lowest weed seed density. Contrarily, chisel plough was recorded with statistically minimum ($p \leq 0.05$) weed seed density (2,599 seeds m^{-2}) at the 20 25 cm soil layer.

Data recorded in 2014 reflects non-significance of weed management practices throughout the soil cores (0-25 cm). Tillage systems were only found to be statistically significant in modulating weed seed density. Soil samples taken at the 0-5 cm depth with the cultivator, rotavator and chisel plough treatment were recorded with statistically maximum weed seed

Table 1 **-** Presence and absence of weed species in weed flora and in soil weed seed bank throughout 2013 and 2014

Scientific name	Common Name	T_1 (1)			T_2			T_3			T_4		
		W_1 (2)	W_2	W_3	W_1	W_2	W_3	W_1	W_2	W_3	W_1	W_2	W_3
Trianthema portulacastrum	Horse purslane	W-S (3)	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S
Echinochloa colona	Jungle rice	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S
Digera muricata	False amaranth	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S	W-S
Sorghum halepense	Johnson grass	W-S	W-S	W-S	S	W-S	W-S	W-S	W-S	S	S	W-S	W-S
Cynodon dactylon	Bermuda grass	W-S	W-S	S	W-S	S	-	W-S	S	W-S	S	w-s	S
Portulaca oleracea	Purslane	S	S	S	S	-	-	W-S	S	-	W-S	S	S
Echinochloa crus-galli	Barnyard grass	W-S	-	S	-	-	W-S	S	S	W-S	W-S	S	-
Dactyloctenium aegyptium	Crowfoot grass	S	S	-	-	S	S	S	S	W-S	S	S	S
Ipomoea aquatica	Water morning glory	S	S	S	S	-	-	-	S	S	S	S	S
Eleusine indica	Goose grass	S	-	S	W-S	-	W-S	S	S	W-S	-	S	-
Brassica spp.	Mustard	S	S	S	S	-	S	-	S	S	S	S	S
Coronopus didymus	Swine cress	S	S	S	-	-	S	S	S	S	S	S	S
Emex spinosa	Lesser jack	-	-	-	S	-	-	S	S	S	-	S	S
Convolvulus arvensis	Field bind weed	-	S	S		-	-	S	S	S	S	-	S
Rumex obtusifolius	Broadleaf dock	S	S	-	-	-	S	-	S	S	S	S	S
Chenopodium album	Lambsquarters	S		-	S	S	-	S	S	S	S	S	S

(1) T_1 = Cultivator, T_2 = Rotavator + Cultivator, T_3 = MB plough + Cultivator, T_4 = Chisel plough + Cultivator. (2) W_1 = Atrazine at 1.00 kg a.i. ha^{-1} at 20 DAS, W_2 = Hand Weeding at 20 DAS and 40 DAS, W_3 = No Weeding. (3) Presence/absence of weed species, where W = weed flora, S = soil seed bank, W-S = present in both weed flora and soil seed bank.

Table 2 **-** Total weed seed density (m^{-2}) up to 25 cm depth during both years. The data show the means (standard deviations)

Treatment		2013					2014				
		0-5 cm	5-10 cm	10-15 cm	15-20 cm	20-25 cm	0-5 cm	5-10 cm	10-15 cm	15-20 cm	20-25 cm
Cultivator	W_1	9870 b (349)	8059 (2093)	5830 (1600)	5069 (928)	3405 (485)	9615 (1071)	7668 (2184)	5465 (877)	4734 (1106)	2922 (365)
	W_2	9038 c (997)	8564 (1909)	5406 (911)	4695 (362)	3302 (465)	8380 (1678)	8709 (1398)	5032 (677)	4439 (504)	2732 (221)
	W_3	13852 a (1903)	8984 (1447)	4975 (449)	4204 (685)	3332 (548)	15372 (1512)	8736 (1479)	4891 (899)	3863 (386)	3010 (268)
	Mean (T_1)	10920 A	8536 B	5404 C	4656 B	3346 A	11122 A	8371 A	5129 B	4346 B	2888 A
Rotavator	W_1	10142 b (1039)	9172 (577)	6412 (1452)	4740 (641)	2979 (179)	9854 (994)	9501 (1357)	6415 (1069)	4822 (614)	2747 (735)
	W_2	8710 c (654)	8850 (466)	6490 (1242)	4728 (399)	3419 (201)	8484 (625)	8797 (796)	6342 (900)	4690 (748)	2762 (307)
	W_3	10784 b (1521)	9172 (176)	5639 (636)	4406 (1205)	3653 (483)	16117 (285)	9409 (705)	5743 (458)	4223 (685)	3346 (443)
	Mean (T_2)	9878 A	9064 A	6180 B	4625 B	3350 A	11485 A	9236 A	6166 AB	4578 B	2952 A
Mouldboard Plough	W_1	6488 cd (614)	6829 (998)	6940 (916)	5515 (1570)	3358 (532)	6488 (1150)	5922 (554)	7379 (1070)	5801 (1675)	3200 (730)
	W_2	5187 cd (523)	7535 (800)	6426 (826)	5545 (2041)	2991 (963)	4702 (754)	6780 (1027)	6926 (949)	5874 (924)	2952 (595)
	W_3	10594 b (141)	7546 (1082)	7501 (1572)	6868 (920)	3317 (460)	12828 (2299)	6612 (821)	7949 (1417)	6649 (990)	3010 (483)
	Mean (T_3)	7423 B	7304 C	6955 A	5976 A	3222 A	8006 B	6438 B	7418 A	6108 A	3054 A
Chisel Plough	W_1	10696 b (1106)	8442 (675)	5415 (725)	4802 (420)	2692 (394)	10385 (1059)	8204 (1119)	5085 (804)	4486 (953)	2177 (398)
	W_2	8756 bc (1333)	8764 (359)	5699 (1227)	4575 (708)	2845 (487)	8664 (1453)	8836 (1325)	5430 (666)	4166 (381)	2309 (282)
	W_3	13019 a (1193)	9141 (1124)	5626 (1087)	4536 (322)	2258 (255)	17213 (225)	9245 (1532)	5231 (1110)	4048 (1027)	1987 (221)
	Mean (T_4)	10824 A	8782 AB	5580 BC	4637 B	2599 B	12087 A	8761 A	5249 B	4233 B	2158 B
LSD ($p \leq 0.05$)	Tillage	1610	526	664	1021	405	1328	960	1335	917	660
	Weeding	830	-	-	-	-	1063	-	-	-	-
	T × W	1659	-	-	-	-	-	-	-	-	-

Abbreviations: T_1 = Cultivator, T_2 = Rotavator + Cultivator, T_3 =MB plough + Cultivator, T_4 = Chisel plough + Cultivator. W_1 = Atrazine at 1.00 kg a.i. ha^{-1} at 20 DAS, W_2 = Hand Weeding at 20 DAS and 40 DAS, W_3 = No Weeding. Means within a column and sharing the same letter do not differ at the 0.05 signiûcance level.

density (>10,000 seeds m^{-2}). The lowest weed seed density was found in the case of MB plough (8,006 seeds m^{-2}). It inverted the soil to a great extent hence the lowest weed seed density was found at 0-10 cm (6,438 seeds m^{-2}) and the highest weed seed density at 10-20 cm (7,418 seeds m^{-2}) was recorded in the soil samples taken from MB plough treated plots. MB plough caused 25-33% addition in weed seed bank by inverting soil at the 5-10 and 10-15 cm soil layers.

There was modulation of actual weed seeds in this study rather than plastic and ceramics beads used by some other researchers (Mohler et al., 2006) for several reasons. Firstly, plastic beads might cling together because of cohesion. Secondly, beads are round but while some of the weed species produce round seeds, others produce flat and oblong seeds. The weed species found in this study were common in maize fields in Faisalabad. Riaz et al. (2007) reported these species (horse purslane, johnson grass, false amaranth, Jungle rice and bermuda grass) as the main weeds in maize fields of Pakistan. The higher relative abundance of weed seeds of horse purslane and jungle rice was due to their dominance in the weed flora of the field. They were the most abundant with 100% frequency in both weed flora and soil weed seed bank. Hashim and Marwat (2002) also found 16 different weed species in maize in their case study. Not surprisingly, seeds of all the weeds of the weed flora were present in the soil weed seed bank. Upper soil layers had more seeds from seed rain of the previous year.

There was a significant effect of tillage and depth for total weed seed density in both years. Density of weed seeds at the upper soil layer (0-5 cm) was four times as high as density at the 20-25 cm soil depth (Auškalnienõ and Auškalnis, 2009). When the field is plowed using shallower tillage implements, they just stir the soil making the upper layer populated by seeds. Seed density was low in soil layers plowed by MB plough. This decrease was attributed to the inverting action of the MB plough (Rahman et al., 2000). It has buried the surface seeds to the depth while soil from the deeper cores was brought to the upper layer which had lower seed density. Mohler et al. (2006) observed that 97% of the seeds in 4 cm of the soil profile had come from the deeper soil layers by using MB plough. There was a slight decrease in the weed seed bank in the deeper layer tilled with a cultivator and a rotavator. MB plough also incorporated a great deal of air in the soil. When seed bank was recorded after harvesting, there was a decrease in the weed seed bank. That decrease might be due to soil settling after irrigation. The chisel plough alone tapped soil core at 20-25 cm depth and was able to stir and modify weed seed density. This change was attributed to the effective depth of chisel plough as it goes deeper than all other tillage implements used in this study. It might have stirred and brought seeds to upper soil cores. MB plough was significant in burying the seeds to the 20 cm depth more than bringing the weed seeds to the surface. More seeds were concentrated in the 15 20 cm layer of the MB plough treatment, indicating that the MB plough buries some seeds at this depth when inverting the soil. Soil inversion has brought such dynamics for soil weed seed population (Rahman et al., 2000; Vasileiadis et al., 2007). The cultivator and the rotavator were the least effective in changing weed seed density of the soil. Hand hoeing and chemical weed control not only controlled weeds of the field but also reduced deposits to the weed seed bank of the soil by killing weed seedlings prior to seed setting. Different tillage systems lead to changes in both the above-ground weed flora and soil weed seed banks. MB plough in combination with hand hoeing was the best weed seed bank management practice in terms of reducing weed seed density.

Addition to the weed seed density in the upper soil layer was up to 40% in the case of no weeding. Weeds remained unchecked throughout the growing season and added to the existing weed seed bank after seeding. However, in soil layers below 5 cm (5-25 cm), the reduction in weed seed bank was not remarkable. The soil core of 15-20 cm with MB plough as tillage system showed maximum weed seed density (5,976 seed m^{-2}) while the other three tillage systems were at par with each other with lower weed seed density. This increment in weed seed density in MB plough treated plot was linked to the burying of the upper surface soil with high seed density; it was brought to the deeper layer by the inversion of soil. Data recorded in 2014 reflects non-significance of weed management practices throughout the soil cores (0-25 cm). Tllage systems were found to be statistically significant in modulating weed seed density (Barberi and Cascio, 2001). The results of the study can be applied to understand how particular assemblage of weeds can be better managed by tillage and weed management practices. Our data suggests that management practices, such as tillage, weeding and herbicides, act as filters that determine the composition and abundance of weed species in the field.

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