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DO TILLAGE METHODS AFFECT GERMINATION AND SPECIES SIMILARITY OF SOIL WEED SEEDS BANK?

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Cultural practices such as tillage used for crop production influence the composition of the weed seed bank in the soil. In order to investigate the effects of different tillage methods on seed bank properties, species diversity and similarity, two laboratory and greenhouse experiments were carried out as randomized complete block design with four replications in 2011. Treatments included: once tillage per year (T1), twice tillage per year (T2), more than twice tillage (T3) and no tillage (T4). Laboratory results showed that the T3 and T4 treatments had the highest and the lowest observed seeds numbers, respectively. Between the laboratory observed weed seeds, the maximum weed seed numbers were *Echinochloa crus-galli* and *Amaranthus retroflexus* in the T3 treatment, while *Chenopodium album*, *Polygonum aviculare* and *Cuscuta campestris* had the highest seed numbers in the T2 treatment. At the greenhouse study, *Chenopodium album*, *Amaranthus retroflexus* and *Hordeum morinum* in the T2 treatment were dominant species. The highest diversity was observed in the T2 treatment, and *Chenopodium album* and *Echinochloa crus-galli* were dominant species in the T2 and T3 treatments. Maximum species similarity index was achieved from the T1 and T3 treatments. Thereby this study concluded that increasing of tillage number could affect the similarity index of weed seeds and subsequently alters the weed community composition.

Keywords: diversity; germination; Shannon index; Sorenson index; tillage

The compositions of weed communities of agricultural lands vary greatly and are closely linked to cropping history. Species composition and density are influenced by farming practices and vary from field to field and among areas within fields (Buhler et al., 2009). Weed is a plant that interacts with crops, which leads to decreasing of yield quality and quantity. There are a multitude of control methods available to manage weeds. But unfortunately chemical way is the most effective and common manner (Chandramohan et al., 2002). Several negative effects of chemical weed control, however, have been recorded in recent years. Because of negative effects of herbicides on the environment and resistance of weed to herbicides, necessities of non-chemical controlling approaches are being more emphasized. Tillage is often seen as an organic alternative to chemical weed control. Tillage can be very effective in reducing weed populations (Parish, 1990). Tillage practices bring the weed seeds to the top soil layers and expose them to sunlight, resulting in germination, thereby increasing their susceptibility to herbicides and environmental stresses (Buhler, 2002). As some seeds are photoblast, then by preventing of sunlight attaining and decreasing of germination of seeds during the tillage their population could be decreased (XiangJu et al., 2012). Tillage is one of the most important factors in the composition and density of

weeds. Different tillage methods could change the weed flora through the influence of residues on soil seeds germination conditions, humidity and soil temperature changes, and weed seeds dispersion in soil (Baskin and Carol, 1980; Abu-Hamdeh, 2003). Tillage is an important component of integrated weed management. Determination of the maximum germination rate under tillage systems is very important in this sense. Thus, comprehensive information about weed density and weed seeds bank properties is necessary for successful weed management (Buhler, 2002). Field seed bank properties are affected by weed population, thereby depend on cultural methods and management practices (Concenço et al., 2011). Cultural practices such as tillage, rotation and inputs uses are the topics of some researches in weed management science. Cardina and Norquay (1997) showed that annual tillage decreased the trend in the viability of soil seed bank. After 8 years just 5% of primary population were survived and after 17 years it decreased to less than 1%. Blackshaw et al. (1994) reported that different tillage methods affected vertical seeds dispersion in the soil profile. Based on the results of these researchers, the germination and density of weed species was greater in no tillage compared to conventional and minimum tillage. The effect of tillage number on weed seed bank has been reported previously (Buhler et al., 2009; Concenço et al.,

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2011). Therefore, more information is needed to determine the effect of various tillage systems on weed seed bank and seedling characteristics. The objectives of this study were to investigate the effects of tillage operations number on germination and species similarity and diversity and to identify dominant species under different tillage number.

Material and methods

In order to consider the tillage operation number effects on seeds number, germination, species diversity and weed species similarity, two laboratory and greenhouse experiments were carried out at the Faculty of Agriculture, University of Shahrood, Iran in 2011 based on randomized complete block design with four replications. The treatments included once tillage in July (T1), twice tillage in July and October (T2), more than twice tillage in July, October and November (T3) and no tillage (T4). Soil samples were obtained in November. Soil tillage was done by a moldboard plow. Twenty samples per plot were taken at random to determine the seed bank composition. Soil cores, 3.5 cm in diameter, were divided into 0–15 cm depths and the 20 samples from each farm were pooled. In this study, cloth bags were used for separating weed seeds (Forcella et al., 2003). For this purpose, the mesh cloth bags (65 meshes) with dimensions 20 cm × 15 cm were used and fabricated with flame. In addition, these bags also have the strength sufficient for various stages of disassembly. The samples were washed and continued until the particles of sand, gravel and plant debris evacuated. The samples were then dried in an appropriate space in open air conditions. Different species of weed seeds were identified under binocular using different keys (Rahman et al., 1995). Other samples (soil) were transferred to the greenhouse for seed emergence evaluation. The soil was air-dried and sieved through a 2 mm screen to break up large soil clods. The entire sample, minus large rocks and root fragments, was spread in 22 cm square trays and watered twice daily in the greenhouse. Weed seedlings that emerged were identified, counted, and removed. Seedlings of questionable identity were transplanted into pots (with a 20 cm diameter, 25 cm height, and 5 kg capacity) and grown until their identity could be verified. Watering continued for one week after seedling emergence ceased; then, the soil was air-dried, thoroughly mixed, and rewetted to permit further germination. This process was repeated four times or until no more seedlings emerged. The greenhouse thermostat was set to maintain a temperature of 25 °C ±3 °C, with no artificial lighting. Seed bank data (counts of seedlings from the germination procedure plus seeds from the extraction process) and weed density in the field were converted to numbers per m². Seed bank species composition data were used to calculate the indices of species diversity and heterogeneity. The weed species such as *Echinochloa crus-galli*, *Cuscuta campestris*, *Chenopodium album*, *Polygonum aviculare*, *Amaranthus retroflexus*, *Hordeum morinum* and *Aegilops* spp. were found in the experimental field. Diversity index was calculated according to Shannon-Wiener index (1963):

$$H = -\sum \frac{N}{ni} \cdot \log\left(\frac{N}{ni}\right) \quad (1)$$

Species similarity among tillage systems was calculated based on Sorenson index (S) (Magurran, 1988):

$$S = \frac{2W}{(C + J)} \quad (2)$$

Data analysis was performed by the SAS and MSTAT-C softwares. Means comparison was calculated by LSD test at $p \leq 0.05$, and figures were drawn in Excel 2010.

Results and discussion

Laboratory experiment

According to the data analysis (Table 1), tillage number had a meaningful effect on total weed seeds numbers ($p \leq 0.01$). The mean comparison of data showed that the maximum seed number belonged to the more than twice tillage treatment (T3) although this treatment had not significant differences with the twice tillage treatment (Figure 1). Total weed seeds numbers in the T3 treatment were about 17.14%, 69.14% and 90.28% greater than the T2, T1 and T4 treatment, respectively. It seems that by increasing of tillage number weed seed density was increased. Baskin and Carol (1980) reported that spring tillage increased the broadleaf weeds population and decreased the annual narrow leaf weeds compared to no tillage system.

Table 1 Analysis of variance of total weed seed number due to different tillage methods

SV	df	Means squares
		total weed seed number
Block	3	13.39
Treat	3	1386.22**
Error	9	36.24
CV (%)	–	24.66

** Significant at $p \leq 0.01$

Notes: SV – sources of variations; df – degree of freedom; CV – coefficient of variation

Figure 1 Effect of tillage operation number on total weeds seeds number

T1: once tillage per year, T2: twice tillage per year, T3: more than twice tillage, and T4: no tillage

Table 2 Analysis of variance of different weed species seeds number due to different tillage methods

SV	df	<i>Echinochloa crus-galli</i>	<i>Chenopodium album</i>	<i>Cuscuta campestris</i>	<i>Amaranthus retroflexus</i>	<i>Polygonum aviculare</i>	Other weed
Block	3	8.62	0.22	19.97	5.13	6.53	0.28
Treat	3	132.27**	322**	17.98**	8.11**	8.22**	0.27 ^{ns}
CV (%)	–	3.09	1.38	2.21	1.88	2.87	0.51

** and ^{ns}, significant at $p \leq 0.01$ and not significant, respectively

Notes: SV – sources of variations; df – degree of freedom; CV – coefficient of variation

Table 3 Mean comparison of weed species seed number due to different tillage treatments

Treat/weed	<i>Echinochloa crus-galli</i>	<i>Chenopodium album</i>	<i>Cuscuta campestris</i>	<i>Amaranthus retroflexus</i>	<i>Polygonum aviculare</i>	Other weed
Once tillage (T1)	1 b	4.33 b	5.66 a	0.33 b	1.66 a	0.66
Twice tillage (T2)	6.66 b	24.66 a	1.33 ab	3.66 ab	0.33 b	0
More than twice tillage (T3)	16 a	20 b	0.79 b	5.66 a	1.33 a	0
No tillage (T4)	0 b	0 d	0 b	1.33 b	2.33 a	0.33
LSD	0.043	0.02	0.029	0.025	0.039	0.006

Different letters in each column indicate significant difference at $p \leq 0.05$

Results indicate that tillage treatments had a significant effect on *Echinochloa crus-galli* and *Chenopodium album* seeds number while it had non-significant effect on other weed species (Table 2). According to mean comparison results, the highest *Echinochloa crus-galli* and *Amaranthus retroflexus* seeds number was observed in the T3 treatment which had meaningful differences with the T1 and T4 treatments. The lowest weed seeds number was observed in the T4 treatment, except for *Polygonum aviculare*. Weed seeds density in T3 and T2 had an increasing trend compared with the T1 and T4 treatments (Table 3).

Some studies have found that weed numbers increased under zero-tillage (e.g. Arshad et al., 1995; Blackshaw et al., 1994; Derpsch, 2003). Others, however, have found that reducing soil disturbance reduced weed populations. For example, seeding with low-disturbance openers reduced weed numbers in a Saskatchewan study (Oliver et al., 2005), and terminating alfalfa stands without tillage reduced weed numbers in the following crop in Manitoba (Ominski and Entz, 2001). Effects vary with weed type. For example, Blackshaw et al. (1994) found that wild buckwheat and common lambsquarters populations were lower in zero-till, even though total weed numbers were higher in zero-till than conventional till.

In zero-till systems, weed seeds remain on the soil surface. The mortality of these seeds is higher than for seeds buried in the soil. Seed predators, such as mice and insects, feed on some of these seeds, preventing them from entering the soil seed bank. Burying weed seeds with tillage operations increases the number of weed seeds in the soil seed bank and may cause these seeds to persist longer than seeds remained on the soil surface, potentially setting the stage for future weed problems. Banting (1966) found that wild oat seeds buried below a depth of 5 cm remained viable longer than seeds on the soil surface. More recently, Gulden et al. (2003) found that canola seed lost during harvest could enter the state of secondary dormancy and persist for many years in the soil. In this study, the number of persistent seeds found in conventional till plots were higher than in zero-till

plots. Tillage can also be used to deplete the weed seed bank in the soil. This practice involves stimulating weed germination with light tillage, either in spring or fall, and then destroying those weeds.

Greenhouse experiment

Results of the greenhouse experiment analysis of variance (Table 4) revealed that tillage operation number had significant effect on germinated seeds numbers ($p \leq 0.01$). Among the seven identified weeds in laboratory just *Chenopodium album* and *Amaranthus retroflexus* could germinate in greenhouse conditions. No germination in other weed seeds may be related to seeds dormancy (Benvenuti, 2007). A noticeable amount of wild varieties seeds are in primary dormancy during the harvest (Sagar, 1970). A lack of synchronized germination caused by seeds dormancy makes various problems in their control (Benvenuti, 2007). Seeds dormancy in *Chenopodium album* L., *Rumex acetosella* (Baskin and Baskin, 1985), *Polygonum aviculare* (Bouwmeester and Karssen, 1992) and *Avena ludoviciana* (Foley, 2001) had been previously reported.

Table 4 Analysis of variance of total germinated seeds due to different tillage methods

SV	df	Means squares
		total germinated seeds
Block	3	24.39
Treat	3	811.22 **
Error	9	21.84
CV (%)	-	3.55

** Significant at $p \leq 0.01$

Notes: SV – sources of variations; df – degree of freedom; CV – coefficient of variation

As shown in Table 5, the effect of tillage number on all weed species germination was significant ($p \leq 0.01$), except for

Table 5 Analysis of variance of weed species seeds germination due to different tillage operation number

SV	df	<i>Hordeum morinum</i>	<i>Chenopodium album</i>	<i>Amaranthus retroflexus</i>	<i>Aeigilops spp.</i>
Block	3	15.22	4.16	0.22	0.06
Treat	3	413.56**	58.83**	1.22 ^{ns}	0.56**
CV (%)	-	3.44	1.70	0.77	0.25

** and ^{ns}, significant at $p \leq 0.01$ and not significant, respectively

Notes: SV – sources of variations; df – degree of freedom; CV – coefficient of variation

Table 6 Means comparison of weed species seed germination in response of tillage operation number

Treat/weed	<i>Hordeum morinum</i>	<i>Chenopodium album</i>	<i>Amaranthus retroflexus</i>	<i>Aeigilops spp.</i>
Once tillage (T1)	0.75 ^b	0 ^b	1.25 ^a	0 ^b
Twice tillage (T2)	21.25 ^a	7.75 ^a	0.25 ^a	0.75 ^a
More than twice tillage (T3)	2.25 ^b	2.25 ^b	0 ^a	0 ^a
No tillage (T4)	0 ^b	0 ^b	0.25 ^a	0 ^a
LSD	6.79	2.96	1.25	0.39

Different letters in each column indicate significant difference at $p \leq 0.05$

Table 7 Biodiversity indices and species dominant of weed seeds bank in different tillage treatments

	No tillage (T4)	Once tillage (T1)	Twice tillage (T2)	More than twice tillage (T3)
Shannon index	0.88	0.96	1.41	1.16

Amaranthus retroflexus. The comparison of means of data showed (Figure 2) that the maximum seeds germination percentage of *Chenopodium album*,

Hordeum morinum and *Aeigilops spp.* was observed in the T2 treatment which had meaningful differences with other treatments (Table 6). This result was

similar to the laboratory experiment results.

With respect to the results indicated in Table 7, generally Shannon index or species diversity in all tilled treatments was less than 1.44, while the maximum and minimum Shannon diversity index were related to the T2 and no tillage treatments, respectively. On the other hand, less dominant weed number was observed in the no tillage treatment. The abundance of different weed species in different tillage treatments is shown in Figure 3. The dominance of both *Echinochloa crus-galli* and *Chenopodium album* was high in the T2 and T3 treatments, which led to increasing of species diversity.

Tillage can also be used to deplete the weed seed bank in the soil. Increasing of weed germination by increasing of tillage operation number may be related to increasing of soil ventilation and increasing of gas exchange between the seed and environment, scratch of seeds by soil gradient, or machines which led to stimulant germination (Scopel et al., 1994; Benech-Arnold et al., 2000).

In order to investigate the weed seeds bank similarity under different tillage treatments, Sorenson index was used. The similarity matrix index results (Table 8) indicated seeds bank similarity in different tillage systems. According to these results, the highest

Figure 2 The means of total weed seeds germinated in response to different tillage operation number

T1: once tillage per year, T2: twice tillage per year, T3: more than twice tillage, and T4: no tillage

Figure 3 Abundance of weed seeds number under different tillage treatments
T1: once tillage per year, T2: twice tillage per year, T3: more than twice tillage, and T4: no tillage

Table 8 Sorenson species similarity index of weed seed bank under different tillage treatments

	Once tillage (T1)	Twice tillage (T2)	No tillage	More than twice tillage (T3)
Once tillage (T1)	–	0.9	0.5	1.0
Twice tillage (T2)	–	–	0.66	0.9
No tillage	–	–	–	0.5
More than twice tillage (T3)	–	–	–	–

seed bank similarity was observed in once tillage (T1) with more than twice tillage (T3) and twice tillage (T2) with once tillage (T1) treatments, while the lowest was observed in no-tillage with once tillage and no-tillage with more than twice tillage treatments. In comparison of four management systems, Concenço et al. (2011) reported that similarity index was different between no-till and conventional tillage.

Conclusion

The number of soil tillage had a considerable effect on weed seed bank composition. Results showed that germinated seeds percentage in once tillage, no-tillage and more than twice tillage were less than twice tillage (T2). The modification of the soil environment due to ploughing resulted in an increase in the abundance of weed seeds. The maximum diversity was related to the twice tillage treatment and *Echinochloa crus-galli* and *Chenopodium album* were dominant weed species both in the twice and more than twice tillage treatments. It could be concluded that increasing of tillage number could affect the similarity index of weed seeds and subsequently alters the weed community composition in agricultural systems.

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