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The content of weed seeds in the soil based on the management system

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Abstract. In 2008 an experiment was set up on the field in Eerika experimental station (Estonian University of Life Sciences) as a 5-field crop rotation: barley (*Hordeum vulgare* L.) with undersown red clover, red clover (*Trifolium pratense* L.), winter wheat (*Triticum aestivum* L.), pea (*Pisum sativum* L.), potato (*Solanum tuberosum* L.). The objective of the study was to measure the content of weed seeds in the soil and to evaluate the diversity of the species at the beginning of the period of organic production in 2011. In conventional farming systems without fertilizer (Con I) and conventional farming with mineral fertilizer (Con II) herbicides were used for weed control. All the crops in Con II system received P 25 kg ha^{-1} and K 95 kg ha^{-1}, but the application rates of mineral nitrogen fertilizer differed. In organic systems (Org I – organic farming based on winter cover crop and Org II – organic farming based on winter cover crop and manure), the winter cover crops (ryegrass after winter wheat, winter oilseed rape after pea, winter rye after potato) were sown after the harvest and were ploughed into the soil as green manure in spring. Organic farming systems (Org II) had a negative effect on the content of weed seeds in the soil (2.0–22.7% less seeds than in other variants). The seeds of *Chenopodium album* were the most abundant among summer annual weeds and the seeds of *Viola arvensis* among winter weeds in the soil. Organic farming measures increased the domination of *Chenopodium album* – the dominance index *D'* was increased by 0.09–0.14 compared to conventional variants. The content of seeds of winter weed *Viola arvensis* in Org II variant was decreased by 82%. The index of species evenness *J'* and Shannon Wiener diversity index *H'* were lower in organic plots by 0.10–0.18 and 0.60–0.19, respectively. Org II variants showed the best results based on the decrease of soil weed seed bank and distribution of the weed species.

Key words: organic farming, winter cover crops, weed seeds, species diversity.

INTRODUCTION

The weed seed bank consists of many species, whereas some dominant species may comprise 70–90% of the total seed bank causing the most problems (Wilson, 1988). Plant protection practices without the use of herbicides has been pointed out as one of the main problems of successful organic cultivation system (Gianessi & Reigner, 2005). In organic farming weeds are the main crop yield decreasing factors. In ecologically friendly cultivation weeds can be controlled by tilling, crop rotation, harrowing, mulching, growing legumes in rotation and cover crops (Watson et al., 2002; Thorup-Kristensen et al., 2003). Soil movement on no-tilled field may lead to much larger number of emerging weed seeds compared to tilled soils (Chauhan et al., 2006; Tamm et al., 2016). The increase of number of soil weed seeds has also been observed in reduced tillage system (Tørresen et al., 2003; Woźniak & Kwiatkowski, 2012). A vigorous cover crop can change the environment for weed seeds on and in the soil. Previous results indicate, that winter cover crops reduced the dry biomass and density of weeds compared to the conventional control system (Madsen et al., 2015).

The aim of the study is to investigate the influence of different winter cover crops and their combination with composted cattle manure on soil weed seed bank at the start of organic farming. The effect of the yield of winter cover crops and preceding crops on the number of weed seeds in the soil was studied in detail.

MATERIALS AND METHODS

The five-field crop experiment with three different organic systems was started in 2008 and was set up in test site of the Estonian University of Life Sciences in Eerika (58°22′ N, 26°40′ E). A 5-year crop rotation contains: red clover *RC* (*Trifolium pratense* L.), winter wheat (*Triticum aestivum* L.), pea (*Pisum sativum* L.), potato (*Solanum tuberosum* L.) and barley (*Hordeum vulgare* L.) undersown with red clover. First 5-year rotation ended in 2012. The experiment was established in four replications, each plot (60 m^2) situated in a systematic block design (80 plots). Organic and conventional plots were separated with an 18 m long section of mixed grasses to avoid contamination with synthetic pesticides, mineral fertilisers and winter cover crops. In systems Con I (conventional farming without fertilizers, as control) and Con II (conventional farming with mineral fertilizers) herbicides (Roundup 4 L ha^{-1} were used in last autumn). In conventional system Con II all the crops received phosphorous (P 25 kg ha^{-1}) and potassium (K 95 kg ha^{-1}). The amount of nitrogen (N) varied depending on the crop: for pea N 20 kg ha^{-1}, for barley undersown with red clover N 120 kg ha^{-1}, for winter wheat and potato N 150 kg ha^{-1}. Plots with red clover did not receive any mineral fertilizers and chemical pest control. Two organic farming systems (Org I and Org II) were investigated. In both systems winter cover crops as green manure were used. Cover crops were sown right after the harvest: ryegrass – *RG* (*Lolium perenne* L., sowing rate 25 kg ha^{-1}) after winter wheat, winter oilseed rape *WOR* (*Brassica napus* L., *var. oleifera, subvar. biennis*, 7.1 kg ha^{-1}) after pea and winter rye *WR* (*Secale cereale* L., 220 kg ha^{-1}) after potato. Before sowing the subsequent crop all cover crops were ploughed into the soil as green manure. The growth stage (according to BBCH) of cover crops before ploughing was determined (ryegrass 21–22, winter oilseed rape 24–26 and winter rye 23–26. In the organic system Org II fully composted cattle manure was added in the autumn 2010 and in spring 2011 at a total rate of 40 t ha^{-1} and ploughed into soil. The cover crops were sown with the Kongskilde sowing machine right after the harvesting of the main crop and in the beginning of May they were ploughed into the soil. Red clover was cut twice: middle of June and during the second half of July and ploughed into the soil in all organic systems. Cereals and peas were harvested at the beginning of August, using a Sampo Rosenlew experimental harvester. Potato tubers were hand-collected between August and September. Yield data were adjusted to dry matter content. Samples for measuring the weed seed bank were collected in September

2011, at the start of first year to full organic production. The weed seed bank samples were taken with soil borers (15 mm diameter) after crop harvest and before autumn ploughing. From each plot 16 soil samples were taken from the depth of 0–25 cm soil layer. Samples of each plot were mixed together in a bucket. The samples were air-dried and 500 g of each sample were sieved and washed through a 0.25 mm sieve. Weed seeds were separated from the soil by potassium carbonate (K_2CO_3) aqueous solution. For preparation of the solution 2.0 kg of potassium carbonate was dissolved in 1.8 L of water. The number of weed seeds in seed bank was calculated to an area of 1 m^2 of plot area using the formula Eq. 1 (Kuht et al., 2016) as shown below:

$$N = \frac{h \cdot \rho \cdot n \cdot 10}{Wd} \tag{1}$$

where N – number of viable seeds, n m^{-2}; h – depth of plough layer, cm; ρ – soil bulk density, g cm^{-3}; n – counted number of seeds in the soil sample; Wd – weight of dry soil sample, g.

The species composition of weed seed communities and the number of seeds of each species were used to assess the biodiversity.

The diversity indexes (Eq. 2) of weed seed species were calculated as Shannon-Wiener diversity index (Shannon, 1948; Shannon & Wiener, 1949) of weed seed species diversity H':

$$H' = -\sum_{i=1}^{S} pi \ln(pi) \tag{2}$$

The community dominance was determined based on the Simpson index, Eq. 3. Simpsons' index of species dominance (Simpson, 1949) was calculated as follows:

$$\lambda = \sum_{i=1}^{S} pi^2 \tag{3}$$

The formula to calculate Pielou evenness index, Eq. 4 (Pielou, 1966; Boyce, 2005) is as follows:

$$J' = \frac{H'}{H'max} \tag{4}$$

The richness index, Eq. 5, was measured based on the Margalef index (Margalef, 1958) as shown below:

$$R = \frac{S-1}{\ln N} \tag{5}$$

where the pi's are the proportion of all observations in the i^{th} species category; $H'max = \ln(S)$; S – number of species (the total number of weed types in a single treatment); N – total number of individuals of all the species in the block.

The results were analysed by using STATISTICA 7.0: ANOVA, Fisher (LSD) test (Statsoft Inc, 2005). Correlation analysis was used to study the correlation between different number of weed seeds and some indicators of soil physical properties. Linear correlation coefficients between variables were calculated, the significance of coefficients being $P < 0.01$, $P < 0.05$.

RESULTS AND DISCUSSION

As an average, there were significantly less viable annual weed seeds in conventional variants with mineral fertilizers and herbicides (Con II) and organic variants with winter cover crops and cattle manure (Org II) compared to other cultivation methods by 3.1–6.1 and 2.5–5.6 (thousand seeds per 1 m^2) respectively (Table 1). In the soil of Org II variant, there were less seeds (as average) in the plots of potato and barley undersown with clover.

Table 1. Number of annual (summer, winter and total) weed seeds in the soil of different crops in 2011

Crop and preceding crops	Weed seeds	Number of seeds, 1,000 seeds per m^2			
		Con I	Con II	Org I	Org II
Red clover	Summer annual	24.22^{a}	20.58^{a}	28.42^{a}	21.00^{a}
	incl. *Chenopodium album*	19.04^{a}	18.20^{a}	27.16^{a}	19.88^{a}
1) barley, us. r. clover	Winter annual	0.98^{a}	0.70^{a}	1.12^{a}	2.10^{a}
2) potato	incl. *Viola arvensis*	0.14^{a}	0.56^{a}	0.14^{a}	0.28^{a}
3) pea	Total	25.20^{a}	21.28^{a}	29.54^{a}	23.10^{a}
Winter wheat	Summer annual	25.06^{b}	20.30^{a}	20.16^{a}	20.44^{a}
	incl. *Chenopodium album*	19.88^{a}	17.78^{a}	19.32^{a}	19.60^{a}
1) red clover	Winter annual	1.54^{a}	2.80^{ab}	1.82^{a}	3.78^{b}
2) barley, us. r. clover	incl. *Viola arvensis*	0.84^{a}	2.38^{b}	0.70^{a}	0.70^{a}
3) potato	Total	26.60^{b}	23.10^{a}	21.98^{a}	24.22^{ab}
Pea	Summer annual	27.72^{b}	20.02^{a}	20.30^{a}	23.24^{ab}
	incl. *Chenopodium album*	25.20^{a}	17.78^{a}	19.74^{a}	22.82^{a}
1) winter wheat	Winter annual	4.90^{b}	3.36^{ab}	1.68^{a}	1.54^{a}
2) red clover	incl. *Viola arvensis*	4.62^{a}	2.52^{a}	0.42^{a}	0.28^{a}
3) barley, us. r. clover	Total	32.62^{b}	23.38^{a}	21.98^{a}	24.78^{ab}
Potato	Summer annual	20.80^{a}	19.24^{a}	21.71^{a}	15.73^{a}
	incl. *Chenopodium album*	18.46^{a}	18.98^{a}	20.80^{a}	14.82^{a}
1) pea	Winter annual	0.52^{a}	3.38^{b}	1.30^{a}	1.56^{ab}
2) winter wheat	incl. *Viola arvensis*	0.52^{a}	2.47^{b}	0.65^{a}	0.26^{a}
3) red clover	Total	21.32^{a}	22.62^{a}	23.10^{a}	17.29^{a}
Barley, us. red clover	Summer annual	27.02^{c}	14.84^{a}	21.42^{b}	13.16^{a}
	incl. *Chenopodium album*	24.22^{b}	13.16^{a}	19.60^{b}	12.60^{a}
1) potato	Winter annual	1.40^{a}	0.98^{a}	0.98^{a}	1.12^{a}
2) pea	incl. *Viola arvensis*	0.70^{a}	0.56^{a}	0.14^{a}	0.00^{a}
3) winter wheat	Total	28.42^{c}	15.82^{ab}	22.40^{bc}	14.28^{a}
Average	Summer annual	24.96^{b}	19.00^{a}	22.40^{ab}	18.71^{a}
	incl. *Chenopodium album*	21.36^{b}	17.18^{a}	21.32^{b}	17.94^{ab}
	Winter annual	1.87^{a}	2.24^{a}	1.38^{a}	2.02^{a}
	incl. *Viola arvensis*	1.36^{ab}	1.70^{b}	0.41^{a}	0.30^{a}
	Total	26.83^{b}	21.24^{a}	23.78^{ab}	20.73^{a}

Note. Within the same row, values with different letters are significantly different (ANOVA, Fisher (LSD) test); 1) – preceding crops in 2010; 2) – preceding crops in 2009; 3) – preceding crops in 2008.

There were a lot more (11 times more as an average) weed seeds of summer annuals than winter weeds, dominated by the seeds of common lambsquarter (*Chenopodium album* L.). The domination of the seeds of common lambsquarter in various cultivation systems has been observed by many authors (Caroca et al., 2011). It was found that the proportion of the seeds of common lambsquarter among summer annuals was significantly higher in organic plots than in conventional variants. It was 95.2% in Org I, 95.9% in Org II and 85.6% and 90.4% in Con I and Con II systems (4.8–10.3% more in organic systems). Except the Con II variant with potato, the domination of the seeds of common lambsquarter was apparent in all the organic variants.

Among winter annual weeds the most dominant species was field pansy (*Viola arvensis* Murr) and as an average they were present in significantly smaller numbers in organic variants compared to the conventional plots (Table 1). On the contrary to the seeds of common lambsquarter which share among summer annuals was higher in organic variants, the proportion of field pansy seeds in summer annuals in Org I and Org II treatments was 0–50.0% in winter annuals (50.0–100% in conventional plots).

The yield of preceding crop proved to be one of the main factors influencing the number of weed seeds. Clear negative correlation ($r = 0.56$) between dry matter yield of crops (without the clover) and the number of weed seeds was observed in 2010 (Fig. 1). Moreover, very strong relationship ($r = 0.83$) between them was apparent in Org II and Con II systems. It was found that with higher dry matter yield of a preceding crop there were less weed seeds in the soil. For most of the crops studied, weed densities were lower when a crop was grown in rotation (Liebman & Dyck, 1993).

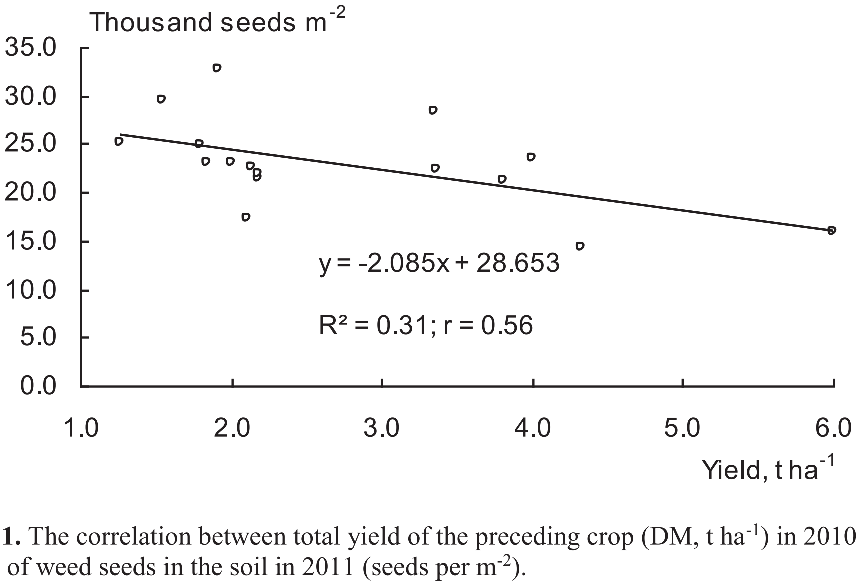

Figure 1. The correlation between total yield of the preceding crop (DM, t ha^{-1}) in 2010 and the number of weed seeds in the soil in 2011 (seeds per m^{-2}).

The correlation between dry matter yield of Org I variants and the number of weed seeds was $r = 64$, but there was no significant relationship with the yield of Con I system. In 2011, the correlation between yields and the number of weed seeds was $r = 0.32$. The number of weed seeds was mostly affected by winter rye (sown as green manure for barley preceding crop and ploughed into the soil in spring) and ryegrass (ploughed into the soil before sowing the pea) the reduction in the number of weed seeds by 17.2% and 16.4% respectively (Fig. 2). Similar effect of reducing the density of weed plants was seen in the same plots where winter cover crops were used. Also, Miura & Watanabe (2002) found that living mulch is effective for weed control and Liebman & Dyck (1993) concluded that weed biomass was lower in the living mulch system. The use of cover crops has an effect on the germination and sprouting of weed seeds, by influencing the light conditions, soil temperature and soil moisture (Teasdale, 1996). Weed presence was strongly influenced by green manure crop, rye being the most suppressive one and ryegrass the least. This had a carry-over effect on weed density in the next pea crop (Barberi et al., 2014). Davis et al. (2005) observed that weed seed banks in the organic systems were positively correlated with weed biomass whereas seed banks in the conventional systems were less predictive. The small reduction (7.2%) of weed seeds in organic variants with red clover was probably due to the effect of the cover crops sown after the preceding pea and potato crops. The effect of growing potato and red clover on the weeds and their seeds was similar in conventional and organic systems.

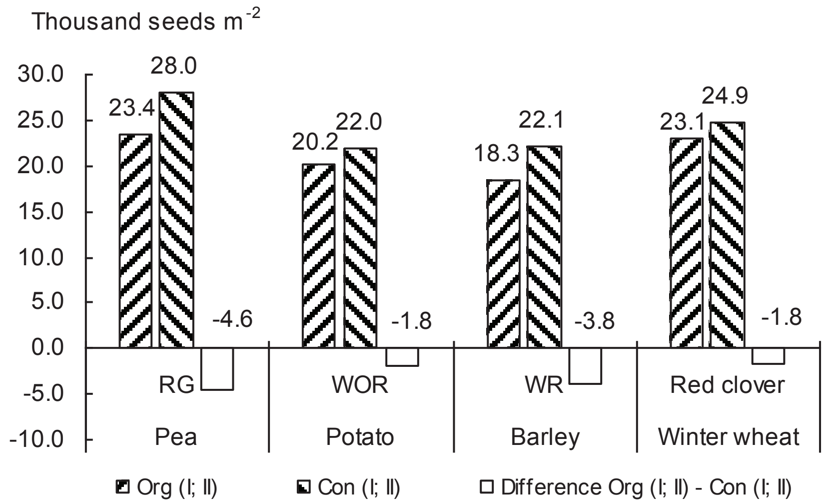

Figure 2. The number of weed seeds in the soil in 2011, depending on the winter cover crops or red clover sown in 2010. *Note*. RG – ryegrass; WOR – winter oilseed rape; WR – winter rye.

In potato plots it was because of the mechanical weeding and in clover plots it was due to the dense canopy that competed with annual weeds during the whole growing period. Also the red clover undersown to barley reduced the density of weeds. Albrecht (2005) also found that the soil weed seed bank was reduced by 39.0% when grasses undersown with clover were used.

Cover crops increase destruction of weed seed in fields. Additionally the increase in the activity of weed seed predator insects was observed. Our results showed that the increase in number of omnivorous predators was observed in areas where winter cover crops had been cultivated previously (Org I and Org II systems, Kuht et al., 2016). The dominant species was *Harpalus rufipes,* that was significantly more present on all organic plots compared to conventional ones. Previous studies have not revealed any significant changes in the number of seed beetles between crops. In our present experiment the *Harpalus rufipes* was significantly more present in pea treatments compared to other crops. As an average of 2 year experiment, diversity index of seed beetles (species per area) was higher by (35%) in organic systems with winter cover crops, compared to the conventional treatments. This is partly due to the better wintering conditions for seed beetles offered by winter cover crops. Blubaugh et al. (2016) also indicated that the cultivation of cover crops increased the activity of weed seed eating insects (omnivorous predators) on the field by enabling their activity on the individual level.

As an average, the Shannon-Wiener diversity indexes (*H'*) were lower in organic variants than in conventional variants (by 0.06–0.19, Table 2), while the largest reduction (by 0.32–0.44) of species diversity was seen in weed seeds in the pea variant.

Simpson dominance indexes (λ) were higher in organic variants than in conventional variants (by 0.09–0.14), especially in the plots with pea. It was obvious that higher dominance index was due to the increase in the share of common lambsquarter seeds in the soil of organic systems.

Table 2. Species diversity index, dominance index and richness index of the weed seed bank in different crops in 2011

Crops	Shannon-Wiener diversity index, *H'*				Simpson's dominance index, λ				Margalef richness index, *R*			
	Con I	Con II	Org I	Org II	ConI	Con II	Org I	Org II	ConI	Con II	Org I	Org II
Barley	0.65	0.68	0.48	0.66	0.60	0.61	0.78	0.66	0.39	0.31	0.60	0.42
R. clover	0.67	0.49	0.30	0.60	0.61	0.74	0.85	0.74	0.39	0.30	0.39	0.70
W. wheat	0.49	0.56	0.56	0.47	0.73	0.70	0.77	0.78	0.20	0.30	0.40	0.40
Potato	0.48	0.62	0.46	0.67	0.76	0.72	0.82	0.74	0.30	0.60	0.60	0.82
Pea	0.73	0.79	0.41	0.35	0.62	0.60	0.81	0.85	0.38	0.50	0.60	0.40
Average	0.61	0.63	0.44	0.55	0.66	0.67	0.80	0.76	0.33	0.40	0.52	0.55

The values of Margalef species richness index (*R*) were higher in organic variants (by 0.12–0.22) compared to conventional variants. Higher species richness indexes in organic variants were observed in plots with red clover and potato. The reason for this here lies also in the agrotechnological characteristics where the effect on the weeds is expressed by intensive mechanical weeding and manure applied with potato and dense canopy and manure applied to the preceding barley crop with red clover.

As an average the evenness index values of Org I and Org II weed seeds were lower than respective values for conventional systems: by 0.18 and 0.12, compared to Con I and by 0.16 and 0.10, compared to Con II variants. The changes in the evenness indexes appeared more clearly in the experiment than species richness and dominance indexes and these are presented in Fig. 3.

The lowest values for evenness index were observed in Org II and the highest values in control variant Con I (except for winter wheat plot), whereas the biggest difference (by 0.24) between conventional and organic systems was seen in plots with pea. The results of correlation analysis indicated that as an average there was a strong relationship ($r = 0.87$) between dominance and evenness indexes, while very strong relationships were seen with pea ($r = 0.98$), barley ($r = 0.93$) and clover ($r = 0.96$).

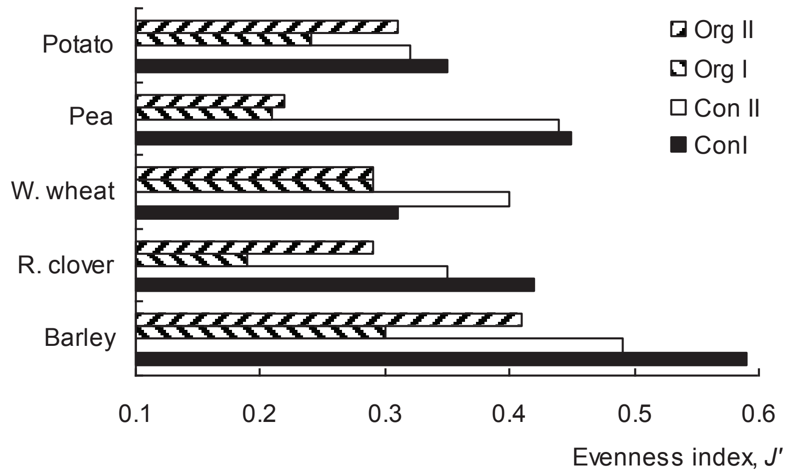

Figure 3. The values of evenness index (*J'*) of weed seeds in the soil of various crops depending on the cultivation system in 2011.

CONCLUSIONS

The yield of preceding crops and cultivation of post-harvest winter cover crops proved to be strong factors for reducing the number of weed seeds in the soil. There were significantly less viable annual weed seeds in conventional variants with mineral fertilizers and herbicides (Con II) and organic variants with winter cover crops and cattle manure (Org II) compared to other cultivation methods. In the soil of Org II variant as an average less weed seeds were found in the soils of potato and barley plots undersown with clover. Common lambsquarter in summer annuals and field pansy in winter annuals were the dominant species. Clear relationship ($r = 0.56$; $P < 0.05$) was found between the dry matter yield of crops in 2010 and the number of weed seeds in the soil in 2011, whereas very strong correlation was seen between Org II and Con II variants. The number of weed seeds was mostly affected by winter rye (sown as green manure for barley preceding crop and ploughed into the soil in spring) and ryegrass (ploughed into the soil before sowing the pea) the reduction in the number of weed seeds by 17.2% and 16.4% respectively. As an average, the Shannon-Wiener species richness indexes were lower in organic variants than in conventional variants (by 0.06–0.19). The lowest values for evenness index were observed in Org II and the highest values in control variant Con I (except for winter wheat plot), whereas the biggest difference (by 0.24) between conventional and organic systems was seen in plots with pea. The values of dominance and species richness index increased in organic variants compared to the conventional variants.

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