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The assemblages of soil-dwelling springtails (Collembola) in winter rye under long-term monoculture and crop rotation

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Abstract

Focusing on the response of soil mesofauna to winter rye cultivation in monoculture for 90 years, the study investigated the abundance and diversity of soil-dwelling springtails, in relation to a five-field crop rotation. A long-term monoculture experiment was established in Poland in 1923 and has been continued uninterruptedly to the present time. Soil samples were taken over a period of three years (2011–2013) to determine springtail (Collembola) abundance, species composition and life-forms spectra. We hypothesize that the crop rotation indirectly, due to the residues from the previous crops and modified soil conditions, caused the increase of the abundance and species diversity of springtails in comparison to monoculture. Generally, springtails were more abundant in the soil of rye field cultivated within a 5-field crop rotation. For the majority of sampling dates the abundance of springtails was higher in that habitat in comparison to the monoculture. Also, the calculated ecological indices showed greater diversity in the crop rotation. This treatment was dominated by the three species – *Hypogastrura manubrialis*, *Mesaphorura macrochaeta* and *Ceratophysella denticulata*, but in the monoculture only *M. macrochaeta* was distinctly numerous. Hemiedaphic springtails dominated in the crop rotation, whereas in the monoculture the most abundant were euedaphic representatives. The results of principal component analysis (PCA) indicated that the studied organisms were influenced by two factors – treatment and the sampling date. We suspect that the unbeneficial soil conditions in certain sampling dates had greater impact on Collembola community than the crop rotation system.

Key words: cereals, Collembola, crop rotation, soil biodiversity, soil mesofauna.

Introduction

Production of cereals constitutes one of the main directions of agricultural production in Poland, and in many other European countries nowadays. As regards the structure of the sown area, cereals make up more than 70% of the total sown area of the country (Eurostat, 2014). The production of such crops, particularly in monoculture or in short rotations, can substantially reduce soil quality and health. Monoculture farming system changes the microclimate, reduces soil porosity, and significantly impacts soil-inhabiting organisms by favouring some groups over others (Rusek, 1998; Brennan et al., 2006; Römbke et al., 2009; Twardowski, 2010). According to Liu et al. (2006), cereals monoculture significantly decreases microbial activity in comparison to short rotation systems. However, there is no reliable evidence how a long-term monoculture affects the soil mesofauna.

In the soil ecosystem life is concentrated especially in a porous space that usually constitutes from 30% to more than 60% of the soil volume in the upper horizons (Lavelle, 2012). This air-filled soil pores are inhabited mainly by soil mesofauna. The most abundant groups are Collembola and Acari, accounting for about 95% of the number of individuals of the total soil arthropod's biota (Nehrer, Barbercheck, 1999).

They play an important role in litter decomposition due to grazing soil fungi and in stimulating nutrient cycling (Rusek, 1998). Springtails respond relatively quickly to any changes in soil environment, so they are commonly used as bioindicators of soil health (Santorufu et al., 2012). Collembola occupy all of the trophic levels in belowground detritus food-webs, but the majority of species feed on decaying organic matter, fungi, and algae. Only a few species feed on plants or are predators (Rusek, 1998; Fiera, 2014). Other results have indicated that Collembola are able to suppress pathogenic nematodes (Stirling, 2014). Springtails occupying different soil subhorizons are classified into life-form groups (epedaphic, hemiedaphic and euedaphic). The distribution of springtails into life-form groups is a routine practice in the studies of the ecological structure of soil collembolan assemblages and can be useful in the monitoring of the soil environment (Sterzyńska, 2009).

In this three-year study, the abundance and species diversity of Collembola were investigated within a 90-year-old monoculture and 5-field crop rotation of winter rye. The low plant diversity (particularly in monocultures) reduces the fauna diversity (Malezieux et al., 2009). Considering the soil functions, it is not proven, that the plant diversity influences processes

occurring in the soil. Under the same soil temperature and moisture conditions, the organic matter will decompose at the same rate under a monoculture as under mixed plants (Kätterer, André, 2009).

In a crop rotation there is a relatively greater diversity of residues from the previous crop plants than in monoculture. We presume that the diversity of crop residues may increase microflora abundance, which may constitute a food source for springtails. Besides, increased amount of organic matter physically changes soil conditions (Bogužas et al., 2015). The results of some studies have showed that the microflora was relatively more abundant in the samples from the crop rotations that included a mix of legumes and cereals (Dorr de Quadros et al., 2012). Going forward, we can hypothesize that crop rotation indirectly increases the abundance and species diversity of springtails in comparison to monoculture, due to the differentiated residues from the previous crops. Especially hemiedaphic springtails, predominantly feeding on microorganisms (secondary decomposers) (Chahartaghi et al., 2005), are able to benefit from the differentiated crop residues in crop rotation.

Material and methods

Experimental site. The experimental fields were located at the Experimental Station in Skierniewice, in the central part of Poland, belonging to the faculty of Agriculture and Biology of Warsaw University of Life Sciences (51°97' N, 20°16' E). A long-term static fertilization experiment was established in Skierniewice in 1923, and has been continued uninterruptedly to the present time. The climate of the study area is transitional, between maritime and continental, with annual mean rainfall of 538 mm (the highest in July and the lowest in February) and mean temperature of +8.6°C (the highest in July and the lowest in January).

The experiment was conducted on winter rye (*Secale cereale* L.) crop, cultivar 'Dankowskie Złote', in 2011–2013. The rye cultivar has not been changed since the experiment was established. Two treatments were used in

the experiment winter rye growing in monoculture and in five-field rotation: potatoes (by 30 t ha⁻¹ of farmyard manure) → barley, → red clover, → winter wheat, → winter rye. In both treatments, the same amount of Ca and NPK mineral fertilizers was applied. Lime (CaO) was applied every fourth year at a rate of 1.6 t ha⁻¹, while N (ammonium nitrate) – 90 kg ha⁻¹, P₂O₅ (superphosphate) – 60 kg ha⁻¹ and K₂O (potassium salt) – 91 kg ha⁻¹ were used every year. Potassium and phosphorus fertilizers were applied in autumn and nitrogen mainly in spring. In monoculture and in rotation, farmyard manure at a rate of 30 t ha⁻¹ was applied in spring every 5th year. In both treatments conventional agricultural practices were used. In autumn it was: disking to a depth of 10 cm, ploughing – 25 cm and harrowing – 5 cm. Before sowing, the following plant protection products were applied, herbicides depending on the year: a.i. iodosulfuron (Huzar Activ 387 OD, 2.4 D acid from 2 EHE and Esteron 600 EC), a.i. 2,4-dichlorophenoxyacetic (Expert Met 56 WG) and a.i. chlorsulfuron (Glean 75 WG). Winter rye was sown in mid-September. The crop was harrowed each year at the end of July.

The experiment was established as a split-plot with five replicates, separately for monoculture and crop rotation. Plants were grown in plots of 36 m² (12 × 3 m). The monoculture plots were arranged in two blocks; the crop rotation plots in three blocks. The plots were randomly arranged within blocks along with other fertilizer treatments. The distance between the blocks was 3 m, with 1 m between the plots. Soil from individual plots was not mixed during the agricultural practices.

Soil conditions. The study was performed on a *Stagnic Luvisol* with clay and silt content in soil layers of about 15% (WRB, 2015). The soil temperature did not show significantly differences among experimental treatments on single sampling dates (Table 1). Since spring 2011 to autumn 2012 there were not noted any significant differences in soil moisture between monoculture and crop rotation. In the spring 2013, the soil moisture was significantly higher in the crop rotation and in the autumn 2013 in the monoculture. Soils in both cropping systems showed pH around neutral (from 6.9 to 7.7 measured in H₂O and 6.0 to 6.9 in KCl).

Table 1. Soil physicochemical properties on sampling dates

Treatment	Spring 2011	Autumn 2011	Spring 2012	Autumn 2012	Spring 2013	Autumn 2013
Soil temperature °C						
Monoculture	13.0 ± 0.2 a	15.1 ± 0.7 a	12.9 ± 0.4 a	13.2 ± 0.6 a	17.2 ± 0.4 a	9.9 ± 0.9 a
Crop rotation	12.8 ± 0.4 a	17.3 ± 0.2 a	11.0 ± 0.5 a	13.4 ± 1.1 a	17.5 ± 0.5 a	9.8 ± 0.8 a
Soil moisture %						
Monoculture	3.8 ± 1.1 a	3.7 ± 0.4 a	11.0 ± 2.7 a	12.5 ± 1.1 a	8.5 ± 1.9 b	12.8 ± 3.4 a
Crop rotation	4.1 ± 0.9 a	4.8 ± 0.7 a	11.3 ± 4.6 a	11.3 ± 1.8 a	13.0 ± 5.4 a	9.7 ± 2.2 b
pH (H ₂ O)						
Monoculture	–	–	7.1 ± 0.05	7.6 ± 0.4 a	7.1 ± 0.6 a	7.0 ± 0.3 a
Crop rotation	–	–	6.9 ± 0.1	7.7 ± 0.3 a	7.4 ± 0.3 a	7.2 ± 0.4 a
pH (KCl)						
Monoculture	–	–	6.4 ± 0.1 a	6.3 ± 0.3 a	6.6 ± 0.43 a	6.9 ± 0.4 a
Crop rotation	–	–	6.0 ± 0.2 b	6.1 ± 0.3 a	6.7 ± 0.4 a	6.4 ± 0.6 a

Sampling. Each year, soil samples were collected twice in spring and twice in autumn. In these seasons an increased activity of mesofauna was observed, which ensured representativeness for quantitative analyses of faunistic material. In spring, soil was sampled

when rye was at the tillering stage (22–27 BBCH) and stem elongation (32–33 BBCH), and in autumn at the germination stage (1–5 BBCH) and leaf development (11–15 BBCH). Each time 25 soil samples were collected from each treatment (5 per plot, across the plot diagonal).

For sampling, a metal core sampler (5 cm diameter, 10 cm depth) with a cutting edge was used. Samples were collected in plastic bags and then transported to the laboratory. Extraction of soil arthropods was conducted in Tullgren funnels modified by Murphy (1956). Each sample was extracted over 24 h. On the basis of our own research, it was found that all springtails leave soil samples within up to 24 h. After extraction, springtails were counted under a stereomicroscope and preserved in 75% ethyl alcohol. Individuals were prepared on permanent slides and identified to the species level under a light microscope on the basis of the following keys (Fjellberg, 2007; Hopkin, 2007). After the determination of the species composition, springtails (Collembola) were attributed to corresponding life forms.

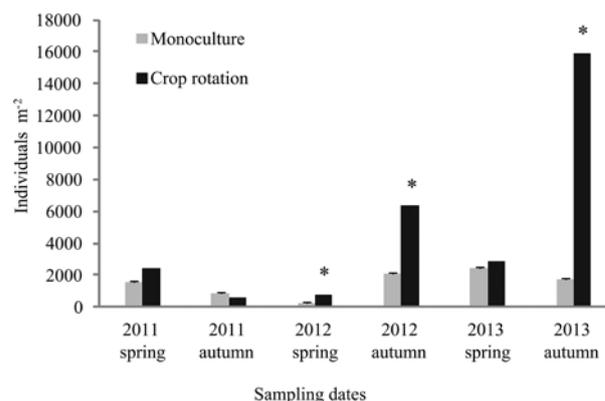
Biological indices and data analysis. For each treatment and season, data on the abundance and diversity of Collembola species were integrated. Total density was analysed by one-way analysis of variance (ANOVA, $p \leq 0.05$) with the use of *Statistica*, version 10.0. For biological calculations Shannon-Weaver, Pielou and Simpson indices were chosen. According to Karaban et al. (2012), all extracted species were attributed to a life form: epedaphic forms (living on the soil surface), hemiedaphic forms (in the upper boundary horizons of litter and soil) and euedaphic forms (in the pore space between particles of soil or sand). Springtails were classified to particular groups based on taxonomical and ecological similarity. Epedaphic forms have strong pigmentation, fully developed furca and appendages, and a complete set of pigmented eyes (8 + 8). Hemiedaphic forms are characterized by the reduction of body pigmentation, eye numbers and a reduced furca. Euedaphic forms have unpigmented body (or eyes' pigmentation), eyes and furca not developed.

Abundance and the community structure of Collembola were calculated separately for spring and autumn. The distribution of Collembola community was analyzed with the ordination method in *Canoco*, version 4.5. Because the gradient length of detrended correspondence analysis (DCA) was less than 2 units, principal component analysis (PCA) was performed (ter Braak, Šmilauer, 2012). As covariables we used treatment and the sampling date. Logarithmic transformation [$\ln(x + 1)$] was performed on species data to normalize the distribution.

Results

Springtails' abundance and diversity. In three of six sampling dates, significantly more collembolan were found in crop rotation than in monoculture (Fig. 1). The highest difference was noted in the autumn 2013, where in all samples collected in crop rotation an average of 15898 collembolans per m² were noted, while in the

monoculture system 1753 individuals per m² were found ($F = 14.0$, $p = 0.0005$). A distinct difference was also observed in the autumn 2012, where significantly more collembolan occurred in crop rotation (6389 m⁻²) than in monoculture (2110 m⁻²) ($F = 12.6$, $p = 0.0006$). Third significant difference was calculated for the spring 2012 when 774.5 m⁻² springtails were recorded in crop rotation system and 275.5 m⁻² in monoculture ($F = 6.3$, $p = 0.01364$). In the autumn 2011, the opposite results were obtained, where springtails were relatively more abundant in monoculture. However, the differences were insignificant.



* – significant differences (ANOVA, $p \leq 0.05$)

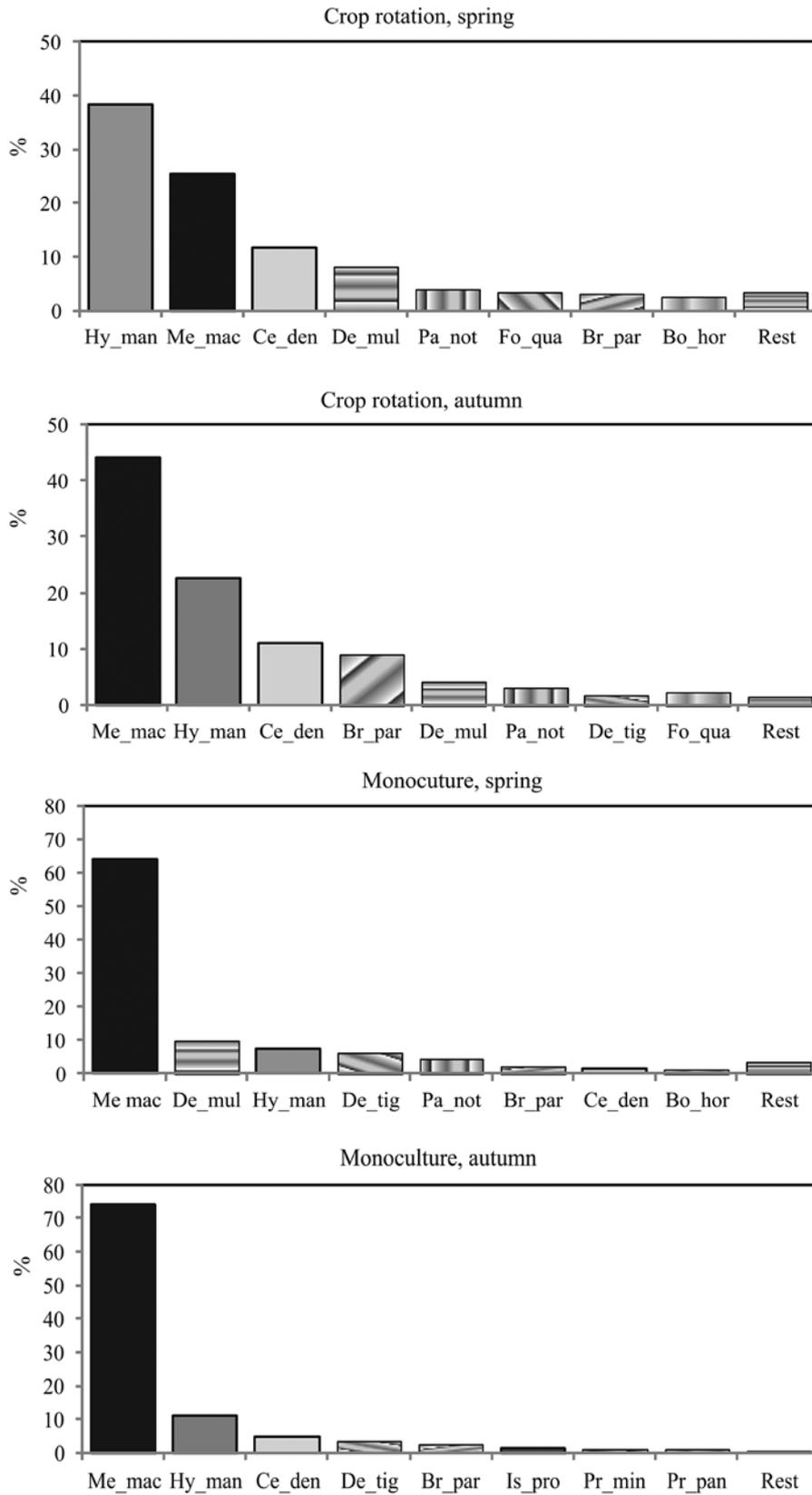
Figure 1. Springtails' abundance in monoculture and crop rotation in 2011–2013

The indices of community diversity, Shannon-Weaver's and Simpson's, in spring in years 2011 and 2013 and in autumn in years 2012 and 2013, indicated higher diversity in crop rotation than in monoculture (Table 2). In the spring 2012 and in the autumn 2011 the diversity indices showed relatively lower diversity in crop rotation. The Pielou value, which takes into consideration the distribution of the abundance of species relative to the total number of species, was higher in crop rotation in spring in years 2011 and 2013 and in autumn in 2013. For other dates, the Pielou value was equal or higher in monoculture than in crop rotation.

In general, the Collembola community was dominated by *Mesophorura macrochaeta* (Rusek) in both studied treatments, contributed from 25.4% to 73.9% of all individuals (Fig. 2). In spring, that species was distinctly numerous in the monoculture system (63.9% of all individuals). Other species constituted less than 10% of all individuals. In crop rotation, three species contributed at least 10% of the total number of Collembola: *Hypogastrura manubrialis* (Tullberg) (38.2%), *M. macrochaeta* (25.4%) and *Ceratophysella denticulata* (Bangall) (11.6%). In autumn, similarly to

Table 2. Springtails diversity in monoculture and crop rotation in spring and autumn

Treatment		Simpson index			Shannon-Weaver			Pielou index			Number of species		
		2011	2012	2013	2011	2012	2013	2011	2012	2013	2011	2012	2013
Monoculture	spring	0.36	0.22	0.54	1.30	1.60	1.04	0.56	0.82	0.45	10	7	10
Crop rotation		0.19	0.28	0.28	1.74	1.59	1.56	0.90	0.73	0.63	7	8	11
Monoculture	autumn	0.29	0.77	0.55	1.32	0.55	0.96	0.82	0.57	0.53	5	7	6
Crop rotation		0.46	0.41	0.29	1.04	1.45	1.48	0.65	0.57	0.59	5	13	12



Me_mac – *Mesaphorura macrochaeta*, Ce_den – *Ceratophysella denticulata*, Hy_man – *Hypogastrura manubrialis*, De_tig – *Desoria tigrina*, De_mul – *Desoria multisetis*, Pa_not – *Parisotoma notabilis* (Schäffer), Br_par – *Brachystomella parvula* (Schäffer), Bo_hor – *Bourietiella hortensis*, Fo_qua – *Folsomia quadrioculata* (Tullberg), Is_pro – *Isotomodes productus* (Axelson), Pr_min – *Proisotoma minuta*, Pr_pan – *Protathorura pannonica* (Haybach)

Figure 2. Frequency of most abundant species of springtails in monoculture and crop rotation in spring and autumn in 2011–2013

spring, *M. macrochaeta* was the most abundant species in monoculture (at least 73.9% of the total number of collembolan). In crop rotation, *M. macrochaeta* (44%), as well as *H. manubrialis* (22.7%) and *C. denticulata* (10.9%) again dominated. Other species that occurred in greater abundance in monoculture in comparison to crop rotation were *Bourietiella hortensis* (Fitch) and *Desoria tigrina* (Nicolet). In contrast, species of the Hypogastrudidae family (*H. manubrialis*, *C. denticulata*) occurred in greater numbers in the crop rotation system, which can be considered as more beneficial for soil-dwelling springtails.

The life form spectra of the Collembola community were similar in spring and autumn (Fig. 3). In monoculture, the most abundant was the euedaphic group (65% and 76%, respectively), then hemiedaphic (23% and 24%, respectively), and finally epedaphic (12% and 0.3%, respectively). In the crop rotation system, the hemiedaphic group occurred in the greatest number (62% in spring and 50% in autumn). Euedaphic forms constituted 27% in spring and 45% in autumn, and epedaphic 11% and 5%, respectively.

Assemblages' structure. The distribution of Collembola species with regard of covariables

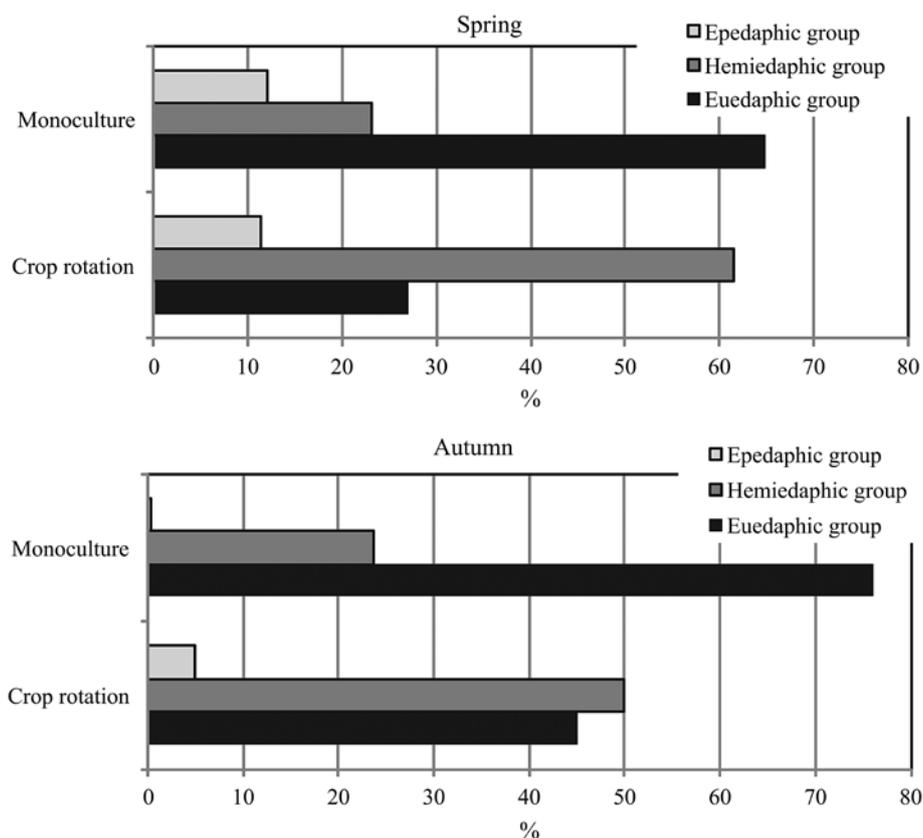


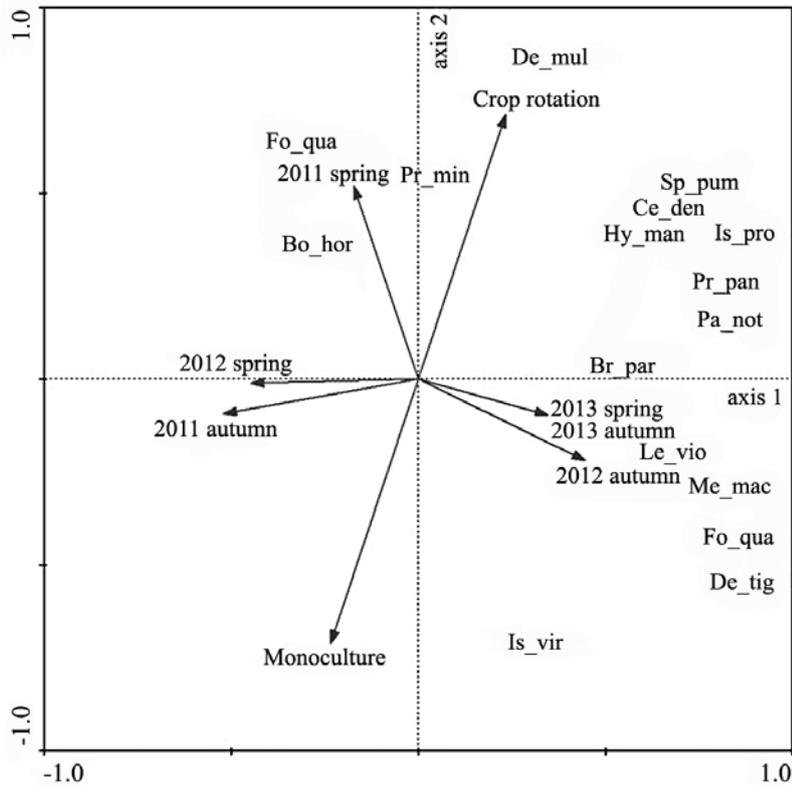
Figure 3. Life form spectra of Collembola in spring and autumn season in 2011–2013

(treatment and sampling date) was shown on the PCA plot (Fig. 4). Analysis of the principal components for Collembola assemblages showed that the first and second component explained 43.7 and 18.7, respectively of the total variance. The first canonical axis was correlated with the most of sampling dates and divided Collembola assemblages into two groups (*Sphaeridia pumilis*, *Ceratophysella denticulata*, *Hypogastrura manubrialis*, and others occurring on the top of the diagram and *Lepidocyrtus violaceus* (Lubbock), *Mesaphorura macrocheta*, *Folsomia quadrioculata* (Tullberg), *Desoria tigrina* occurring on the bottom). PCA analysis placed crop rotation and monoculture on the opposite sides on the diagram and divided springtails into species group occurring on the right side of the ordination plot (most of springtails species) and species on the left side (*Folsomia quadrioculata* and *Bourietiella hortensis*). It was observed, that hemiedaphic species *Proisotoma minuta* (Tullberg) and *Desoria multisetis* (Carpenter & Phillips) were positively clustered with crop rotation.

Discussion

In the studied arable soils, from 275 to 15898 individuals per m² and from 5 to 13 species were found during the study period. Also other authors (Rusek, 1998; Kanal, 2004) reported low densities and species richness of Collembola in arable soils in their studies. In this study, the soil conditions in monoculture and crop rotation (soil temperature, soil pH and soil moisture) did not differ significantly in the particular sampling dates, but they changed distinctly during the experimental period. These changes in soil conditions influenced springtails density and species diversity in particularly seasons. Relatively low Collembola densities were reported in both seasons in 2011 and in the spring 2012. Especially in 2011 this could be the effect of the relatively low soil moisture in mentioned sampling dates. The results of previous studies showed that soil moisture is the critical factor influencing the occurrence of springtails (Devi et al., 2011).

The first part of the hypothesis assuming greater abundance of springtails within more diversified habitat



Me_mac – *Mesaphorura macrochaeta*, De_mul – *Desoria multisetis*, Hy_man – *Hypogastrura manubrialis*, De_tig – *Desoria tigrina*, Pa_not – *Parisotoma notabilis*, Br_par – *Brachystomella parvula*, Ce_den – *Ceratophysella denticulata*, Bo_hor – *Bourietiella hortensis*, Fo_qua – *Folsomia quadrioculata*, Is_pro – *Isotomodes productus*, Is_vir – *Isotoma viridis*, Pr_min – *Proisotoma minuta*, Pr_pan – *Protathorura pannonica*, Le_vio – *Lepidocyrtus violaceus*, Sp_pum – *Sphaeridia pumilis*

Figure 4. The principal component analysis (PCA) ordination biplot of Collembola community with covariables (treatment and sampling date)

was in part confirmed. In half on all sampling dates, the abundance of Collembola was significantly higher in crop rotation in comparison to monoculture. In other dates, no significant differences were noted, but the trend remained. Only in one date (autumn 2011) the opposite results were reported. The results seem to be casual and having no impact on the final conclusion.

Analysing the species diversity, in four of six sampling dates the species diversity was relatively higher in the case of crop rotation. Summarizing the results of other field studies, the effect of crop rotation and monoculture on soil mesofauna is inconclusive. Graenitz and Bauer (2000) indicated higher biological activity of enchytraeids in the monoculture of winter rye in comparison to crop rotation. Jagers op Akkerhuis et al. (1988) found significantly more mites in crops of the six year rotation than in corresponding crops of the three-year rotation, whereas springtails differed not significantly between treatments. Rebek et al. (2002) indicated that the low input cropping system (e.g., grassland) is more beneficial for epedaphic springtails than high management crop (monoculture). We are suggesting that the study results will be more convincing if we correlate the Collembola abundance with the microbial activity.

The diversity of Collembola assemblages did not differ greatly between monoculture and crop rotation. In each treatment the species structure was generally similar. All the species of springtails identified in the

experiment appear to be common and widespread. In monoculture, *Mesaphorura macrochaeta*, which can be considered as a species occurring in large numbers in intensively investigated soils, was most numerous. The distinct domination of *M. macrochaeta* in the monoculture system may indicate the adaptation of this species to relatively unbeneficial soil conditions. *M. macrochaeta* was also the most abundant species in the cereal field in studies conducted by Twardowski (2010).

The life forms spectra in monoculture and crop rotation were differentiated. In monoculture the most abundant were euedaphic springtails, which are most numerous in cyclically ploughed soils (Kanal, 2004). Hemiedaphic springtails, which live near the surface, are more exposed to agricultural treatments, especially ploughing (Chamberlain et al. 2006). As we hypothesized, hemiedaphic springtails were dominant in crop rotation, and after them the euedaphic group. The epedaphic springtails were the least numerous in both experimental treatments. We suspect that this group of springtails is relatively mobile and can even escape during sampling.

Sampling date comparisons were not analyzed statistically because the soil and weather conditions in particular dates were distinctly different. Independently from that, the results of PCA indicated, that the Collembola community was influenced by two factors – treatment and the sampling date. We presume that the unbeneficial soil conditions in certain sampling dates greater affected

Collembola community than the agricultural treatment. Perhaps more intensive sampling (more soil samples and dates) would demonstrate clearer trends in the response of springtail populations to monoculture or crop rotation.

Conclusions

1. In the majority of sampling dates the abundance of springtails (Collembola) was higher in crop rotation in comparison to monoculture.

2. Analysing the species diversity, in almost all the sampling dates the species diversity was relatively higher in crop rotation. This treatment was dominated by three species, i.e. *Hypogastrura manubrialis*, *Mesaphorura macrochaeta* and *Ceratophysella denticulate*. In monoculture, only *M. macrochaeta* was distinctly numerous.

3. Hemiedaphic springtails dominated in crop rotation, whereas in monoculture the most abundant were euedaphic representatives. The results of principal component analysis (PCA) indicated that the studied organisms were influenced by two factors – treatment and the sampling date. We presume that the unbeneficial soil conditions in certain sampling dates greater affected Collembola assemblages than the agricultural treatment.

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Dirvožemio podurų (Collembola) kiekis žieminių rugių monopasėlyje ir sėjomainoje

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Santrauka

Siekiant išsiaiškinti žieminių rugių, 90 metų augintų kaip monokultūra, įtaką dirvožemio mezofaunai, tirtas dirvožemyje gyvenančių podurų (Collembola) gausumas ir įvairovė penkių laukų sėjomainoje. Ilgalakis monokultūros bandymas buvo įrengtas Lenkijoje 1923 m. ir yra tęsiamas iki šiol. Siekiant nustatyti podurų (Collembola) gausą, rūšinę sudėtį ir gyvybės formų įvairovę, dirvožemio ėminiai imti trejus metus (2011–2013). Buvo iškelta hipotezė, kad, palyginus su monokultūra, sėjomaina netiesiogiai, dėl priešsėlių liekanų ir pasikeitusių dirvožemio sąlygų, padidino podurų gausumą ir rūšinę įvairovę. Podurų buvo gausiau dirvožemyje rugių, augintų penkialaukėje sėjomainoje. Daugeliu ėminių paėmimo atvejų podurų buvo gausiau šioje buveinėje, palyginus su monokultūra. Apskaičiuoti ekologiniai rodikliai taip pat rodė didesnę jų įvairovę augalų sėjomainoje. Šiame variante dominavo trys podurų rūšys – *Hypogastrura manubrialis*, *Mesaphorura macrochaeta* ir *Ceratophysella denticulata*, o monokultūroje gausiau nustatyta tik *M. macrochaeta*. Hemiedafinės poduros dominavo sėjomainoje, o monokultūroje buvo daugiausia edafinių. Pagrindinių komponentų analizė parodė, kad tirtiems organizmams turėjo įtakos du veiksniai – variantas ir ėminio paėmimo data. Teigtina, kad podurų kiekiui nepalankios dirvožemio sąlygos kai kuriomis mėginių ėmimo datomis turėjo didesnės įtakos nei sėjomaina.

Reikšminiai žodžiai: Collembola, dirvožemio bioįvairovė, dirvožemio mezofauna, javai, sėjomaina.

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