INFRASTRUKTURA I EKOLOGIA TERENÓW WIEJSKICH
INFRASTRUCTURE AND ECOLOGY OF RURAL AREAS

Nr II/2/2015, POLSKA AKADEMIA NAUK, Oddział w Krakowie, s. 405–417
Komisja Technicznej Infrastruktury Wsi

DOI: http://dx.medra.org/10.14597/infraeco.2015.2.2.033

COMPARISON OF SEASONAL DYNAMICS OF MITE (ACARI) AGGREGATION IN PINE FOREST LITTER AND PINE CHIPS

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Summary

This paper analyzed seasonal dynamics of mite (*Acari*) aggregations, with particular attention to oribatid mites (*Oribatida*), in two different substrates intended for soil regeneration – shredded forest litter from Scots pine forest and pine wood chips. The study was conducted in the years 2011-2012, on microplots established within a belt of trees in a nursery in Białe Błota (Bydgoszcz Forest District). To maintain optimum moisture content, the microplots were hydrated by micro sprinklers as per the guidelines and schedule for the irrigation of nurseries, and mean soil moisture was kept at the level of 5.1-9.9 %. The mites were classified into orders and oribatid mites into species or genera, with regard to juvenile stages. A total of 4,142 mites were determined, including 2,939 oribatid mites.

Mite density in the shredded forest litter, intended mainly for soil inoculation with edaphon, was similar in the initial and final period of the study. Mite density in the pine wood chips, intended for mulching degraded soils and providing optimal conditions for mesofauna development, increased over two years of the study. Oribatid mites were the most abundant mites in the analyzed samples. Mean number of their species *s* in the forest litter was comparable at the beginning and at the end of the study cycle. In the pine chips, the differences in species abundance between the first and second year were significant. Oribatid mites most frequently observed in both substrates included *Chamobates schuetzi*, *Oribatula tibialis* and *Tectocepheus velatus*.

Key words: soil regeneration, mulching, microplots, mesofauna, *Oribatida*.

INTRODUCTION

Colonization of initial soils, featuring harsh conditions for small arthropod survival, is a slow process that may take many months or even years (Beckmann 1988, Lehmitz *et al*. 2011, Wanner and Dunger 2002, Klimek *et al*. 2013a). Therefore, effective methods for improving this process are always looked for. These methods can be used for the regeneration of areas degraded due to various anthropogenic activities.

Reclamation of these areas may be based on such corrective measures as fertilization, liming, renewal of vegetation, organic stimulation of the soil and some bioremediation techniques (Haimi 2000). The aim of these procedures in our climate zone is speeding up forest succession in a way consistent with a type of habitat.

The main objective of the reclamation should be restoring proper functioning of the entire ecosystem. Apart from plants and microorganisms, this process requires also the presence of soil mesofauna. However, this part of edaphon, as not very well understood, is usually overlooked in the remediation activities (Langer *et al*. 1999; Majer *et al*. 2007).

It it worth emphasizing that small soil arthropods, especially the mites of *Oribatida* order, play very important roles in the ecosystems, they improve pedogenic processes and a propagation of bacteria and fungi, and they indirectly affect the formation of endo – and ectomycorrhizas (Klironomos and Kendrick 1996, Behan-Pelletier 1999, Remén *et al.* 2010, Schneider *et al*. 2005). They also serve as good bioindicators of soil biological activity (Behan-Pelletier 1999, 2003, Gulvik 2007). However, these mites have limited possibilities of spreading and colonizing new sites.

Haimi (2000) claims that soil fauna is important for restoring biological activity during the reclamation of degraded soils and that these processes can be reinforced by fauna reintroduction by means of soil inoculum. Our previous studies in nurseries showed that edaphon-rich forest raw humus was very helpful in soil reclamation (Klimek *et al*. 2008, 2012). Particularly good results regarding soil reclamation were achieved in the nurseries of broad-leaf species, i.e. birch and linden (Klimek *et al.* 2013b,c). Due to their quick growth, these species protect the soil against moisture loss and the leaves they shed enrich soil with organic matter.

However, by the time the reclaimed area is adequately covered with vegetation and a layer of raw humus, essential for soil mesofauna development, is formed, the only effective solution supporting reintroduction of these animals seem to be artificial enrichment of soil with organic matter by supplementing the initial soils with compost or mulching. Soil supplementation with compost did not markedly affect the effectiveness of mesofauna reintroduction (Klimek

et al. 2013a,b). Mulching, by imitating a natural structure of forest soil with raw humus layer, seems a better solution. High abundance and diversity of mites in strawberry crop mulched with pine chips was reported previously (Klimek *et al*. 2014a,b). This brought about the idea of using pine chips as a substrate supplementing raw humus during reclamation of degraded forest soils and initiating and shaping forest succession. This is even more promising in the light of the fact that using large quantities of raw humus is limited for reasons of forest soil protection.

The aim of this study was to investigate seasonal dynamics of mite (*Acari*) aggregations, with particular attention to oribatid mites (*Oribatida*), in two different substrates intended for soil regeneration – shredded raw humus from Scots pine forest and pine chips.

(photo by A. Klimek)

Photo 1. Microplots established in a 20 m wide belt of trees in a nursery in Białe Błota

MATERIAL AND METHODS

The study was conducted in the years 2011-2012, on microplots established in a nursery in Białe Błota (Bydgoszcz Forest District). The experiment was established in a 20 m wide belt of trees (53°06'13.2"N, 17°55'46.6"E) in order to mitigate the influence of weather conditions, such as excessive sunlight,

temperature fluctuations or too intensive precipitation. The tree stand included Scots pine (*Pinus sylvestris* L.), oak (*Quercus* L.) and European ash (*Fraxinus excelsior* L.), and the underbush layer was composed of European ash, silver birch (*Betula pendula* Roth) and oak. The soil type was albic brunic arenosol (Bydgoszcz Forest Inspectorate data).

The substrate material – pine forest litter and pine branches (thinning residues) were collected after 11th April 2011. The material was collected near a nursery in a mature Scots pine forest (*Leucobryo-Pinetum* Mat. (1962) 1973) in Białe Błota Forest District. The next day, the collected material was fragmented using a garden shredder VIKING GE 250 and distributed in a 10 cm layer on exposed mineral soil in the designated places within the belt of trees. The experiment involved a total of 4 microplots of 1m^2 each, 2 per each variant: (F) – pine forest litter and (W) – pine wood chips (Pfoto 1). The microplots were isolated from the stand soil by means of 20 cm high Cellfast garden edge inserted at the depth about 10 cm and secured with garden pegs.

To maintain optimum moisture content, the microplots were hydrated by micro sprinklers as per the guidelines and schedule for the irrigation of nurseries, and mean soil moisture was kept at the level of 5.1-9.9 %.

The samples (50 cm^3) for acarologic analyses were collected in the spring, summer and autumn of each study year on the following days: 24th May 2011, 20th July 2011, 27th October 2011, 19th May 2012, 10th July 2012, and 16th October 2012. Ten samples were harvested from each variant (5 from each microplot). A total of 60 samples of 50 cm^3 each were collected from every variant. Mite extraction was carried out over 7 days using Tullgren funnels. Then, the mites were preserved in 70% ethanol. All the mites were classified into orders and oribatid mites into species or genera, with regard to juvenile stages. A total of 4,142 mites were determined, including 2,939 oribatid mites.

Average density (*N*) of these mites was provided for 50 cm^3 of the substrate, and the species dominance index (*D*) was given in percentage. Species diversity was determined based on the mean number of species per sample (*s*). Prior to statistical analysis, the numerical data were subjected to a logarithmic transformation – ln (x+1) (Berthet and Gerard 1965). The statistical analysis was performed using Statistica 6.0, a compliance of the measurable parameters with the normal distribution was assessed using Kolmogorov-Smirnov test. As the normal distribution was not confirmed, a non-parametric analysis of variance (Kruskal-Wallis) was performed. For statistically significant differences ($p<0.05$) a *post-hoc* analysis for each pair was carried out (Mann-Whitney U test) to identify significantly different means (Łomnicki 2000).

RESULTS AND DISCUSSION

Changes in the abundance of mite (*Acari*) aggregations

A successful attempt at soil inoculation with edaphon and raw humus, collected from the mature Scots pine stand (Klimek *et al*. 2013b), was undertaken in Scots pine and birch nursery of Białe Błota. The treatment involved a thin, 1 cm layer of non-shredded raw humus. This form of raw humus slightly hampered the maintenance works during seedling growth. Therefore, in the present experiment, the forest litter was shredded with a garden shredder. At the initial stage of the study, mite density in raw humus substrate was 26.6 individuals per 50 cm^3 (Table 1). After two months, a five-fold increase in the density of these arthropods was observed. However, in the autumn of 2011, the density was reduced to 19.6 individuals per 50 cm^3, and then an over three-fold increase was noticed in the spring of 2012. At the two final sampling dates, the mite density was similar as in the initial period of the study.

Earlier studies have repeatedly demonstrated that forest raw humus is a perfect substrate for soil inoculation with mesofauna (Klimek 2010, Klimek *et al.* 2008, 2011, 2012, 2013b,c). They also showed that inoculation required only a small amount of the substrate. i.e. 1 cm layer was enough. However, in practice, when degraded areas lacking in organic matter are reclaimed, it is necessary to provide more substrate in which the mesofauna would be able to survive and grow. As already mentioned, using greater amounts of forest raw humus is not an option, due to the necessity of renewing the stands after cutting. Hence, the need for an easily available, slowly decomposing organic matter, preferably with a structure similar to the forest litter, providing optimum conditions for mesofauna development. Using wood chips seems to be a good solution to this problem, especially that they can be obtained from logging residues without any environmental costs.

Wood chips are commonly used in gardening for mulching soil surface. Treder *et al.* (2004, 2009) found that wood chip litter used in an apple orchard significantly affected the temperature and humidity changes in the top soil layer and that cooling and heating processes in the soil under the litter were considerably less intense, meaning also better water management in the plants. Soil mulching creates also favorable conditions for a development of microorganisms and small soil fauna (Forge *et al.* 2003). In our previous study, in wood chip mulched strawberry crop, we reported high number of mites – about 20,000 individuals $\cdot$ m^{-2} (Klimek *et al.* 2014a,b). In the first year of the study, the number of mites on the experimental microplots was low (3.5-10.6 individuals per 50 cm^3) but it gradually increased during the study period. In the second year, the number of the arachnids was about two times higher than in the autumn of 2011.

Table 1. Abundance of mites (individuals per 50 cm^{-3}) number species (*S*), average number of species (*s*) of *Oribatida* in (F) – pine forest litter and (W) – wood chips in seasons 2011-2012

Index – Taxon		Research data						Kruskal-Wallis test	
		V. 2011	VII. 2011	X. 2011	V. 2012	VII. 2012	X. 2012	*H*	*p*
N – Acaridida	F	0.1^{A}	0.3^{A}	0	0	0	0	11.59	0.041
	W	0.1	0.1	0.4	0.1	0.4	0.2	2.71	0.745
N – Actinedida	F	4.2^{A}	4.4^{A}	7.7^{A}	12.2^{B}	4.7^{A}	7.2^{A}	12.90	0.024
	W	0.1^{A}	1.5^{B}	1.6^{B}	4.4^{C}	4.8^{C}	6.1^{C}	25.03	0.001
N – Mesostigmata	F	1.4^{A}	12.5^{B}	1.5^{A}	6.1^{C}	3.9^{A}	2.8^{A}	29.33	0.000
	W	3.1	4.5	8.2	4.6	5.6	3.1	11.82	0.374
N – Oribatida	F	20.7^{A}	128.8^{B}	10.3^{A}	48.7^{C}	20.6^{A}	27.2^{A}	37.38	0.000
	W	0.2^{A}	0.8^{A}	0.3^{A}	18.3^{B}	9.7^{C}	8.3^{C}	44.39	0.000
N – Tarsonemida	F	0.2	0.6	0.1	0.2	0.2	0	4.42	0.491
	W	0	0	0.1^{A}	0.1^{A}	0.2^{A}	0.7^{B}	19.21	0.002
N – Acari (Total)	F	26.6^{A}	146.6^{B}	19.6^{A}	67.2^{C}	29.4^{A}	37.2^{A}	36.08	0.000
	W	3.5^{A}	6.9^{AB}	10.6^{B}	27.5^{C}	20.7^{BC}	18.4^{C}	30.89	0.000
S – Oribatida	F	17	16	9	15	16	10	-	-
	W	2	4	2	14	12	12	-	-
s – Oribatida	F	5.20^{A}	7.00^{A}	3.20^{B}	5.40^{A}	4.40^{A}	4.20^{A}	21.56	0.001
	W	0.20^{A}	0.40^{A}	0.30^{A}	4.00^{B}	3.40^{B}	3.50^{B}	43.11	0.000

Explanations: $^{A, B, C}$ – the same letter means the insignificant difference ($p<0.05$)
Source: own research data

On all the sampling dates, the most abundant mites in the pine forest litter substrate were *Oribatida*, accounting for 53-88% of the analyzed populations (Figure 1). The highest density of these mites was reported in the summer of 2011 (Table 1). In the first year of the study, oribatid mites were only occasionally found in pine chips (0.2-0.8 individuals per 50 cm^3). Their density in this experimental variant increased a few times in the second year, and they were the most abundant in the spring of 2012. In 2012, *Oribatida* accounted for 45-67% of all mites in the microplots covered with pine chips, thus becoming the most numerous mites (Figure 1). On the first sampling date, the pine chips were heavily dominated by usually predatory *Mesostigmata* (89% of all *Acari*). This might be due to their size and mobility, as they are usually larger than the other mites and can move quite quickly in their search for food. Their number was relatively similar in both substrates (Table 1). In the forest pine litter, the second group in

terms of abundance and proportion in *Acari* aggregation were usually *Actinedida*. A peak in their number, similarly as in *Oribatida*, was observed in May 2012. In this substrate, the density of *Actinedida*, *Mesostigmata* and *Oribatida* on the first and last sampling date was not significantly different. The mites belonging to *Acaridida* and *Tarsonemida* orders were only occasionally found in the collected material.

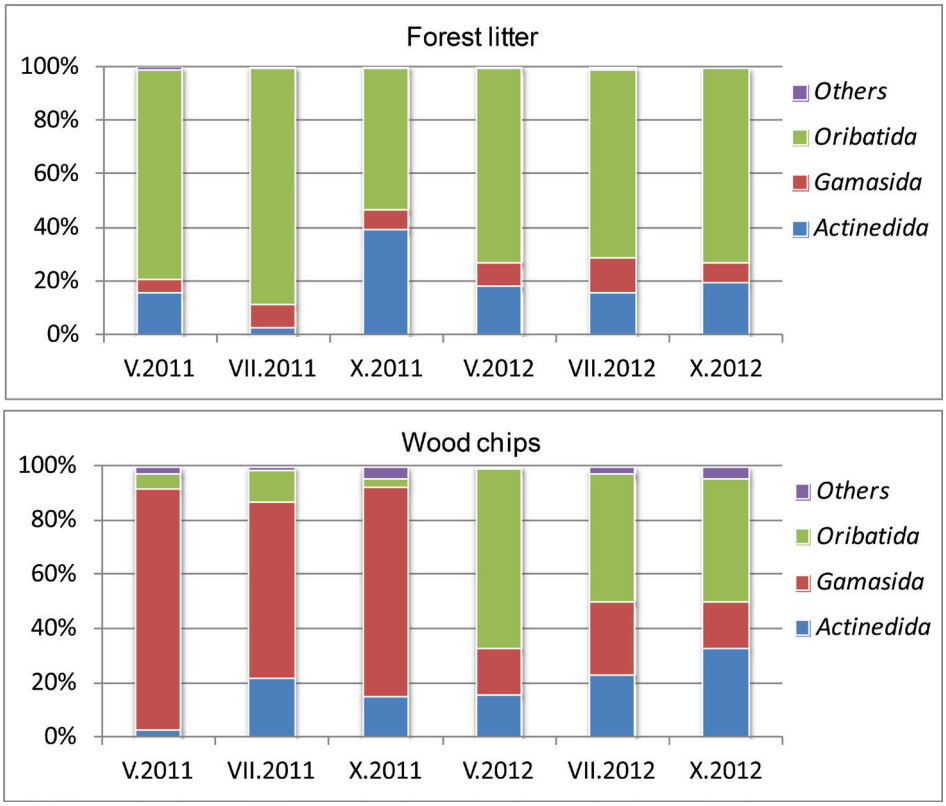

Figure 1. Dominance of taxonomic group of mites in (F) – pine forest litter and (W) – wood chips in seasons 2011-2012

Species diversity of oribatid mites

In total, 32 species of oribatid mites were reported in this experiment, including 28 in the pine litter substrate and 20 in the pine chips (Table 2). In the pine litter substrate, the highest number of *Oribatida* species (17) was observed on the first sampling date, whereas only 10 species were found on the last sampling date. However, a statistical analysis based on average number of species in

a sample (*s*) did not reveal any significant differences between the first and last sampling date (Table 1). A different situation was perceived at the microplots covered with pine chips. At the beginning of the study, only 2 species of *Oribatida* were reported, but in the second year, there were 12-14 species, and the differences between sampling dates in the first and second year of the study were significant.

For example, strawberry crop mulched with pine chips hosted 7 *Oribatida* species at the beginning of a 2-year study, and 11 at the end of this period (Klimek *et al*. 2014b). It is also worth emphasizing that differences in the average number of species *s* between the first and last sampling date were also significant in this case.

Table 2. Abundance of oribatid mites (individuals per 50 cm^3) in (F) – pine forest litter and (W) – wood chips in seasons 2011-2012

Species		Research data						Kruskal-Wallis test	
		V. 2011	VII. 2011	X. 2011	V. 2012	VII. 2012	X. 2012	*H*	*p*
Adoristes ovatus (Koch)	F	0	0	0.1	0.1	0	0	3.11	0.684
	W	0	0	0	0.1	0	0	5.00	0.416
Brachychthonius sp.	F	0	2.8	0	0	0	0	38.77	0.000
Camisia spinifer (C.L. Koch)	F	0.1	0	0	0	0.1	0	4.07	0.540
Carabodes forsslundi Sellnick	F	0	0	0	0.1	0.1	0	4.07	0.540
Carabodes labyrinthicus (Michael)	F	0.2	0.2	0	0	0	0	8.43	0.134
Carabodes subarcticus Trägardh	F	0.3	0.1	0	0	0	0	11.59	0.051
Chamobates schuetzi (Oudemans)	F	8.3^{AB}	4.1^{A}	0.1^{C}	5.8^{A}	4.4^{A}	14.6^{B}	29.52	0.000
	W	0	0.3^{A}	0	0.7^{A}	1.5^{A}	2.6^{B}	14.31	0.014
Cultroribula bicultrata (Berlese)	W	0	0.1	0	0	0	0	5.00	0.416
Damaeus sp.	F	0	0	0.1	0	0	0	5.00	0.416
	W	0	0	0	0.5	0.7	0.3	14.32	0.014
Eremaeus oblongus C.L. Koch	F	0.9	2.0	0.4	1.2	1.0	1.3	2.03	0.846
Eupelops occultus (C.L. Koch)	F	0	0	0	0.1	0	0	5.00	0.416
	W	0	0	0	0.3	0	0	10.17	0.071
Eupelops torulosus (C.L. Koch)	F		0.1	0.2	0.1	0.1	0	2.11	0.831
	W	0	0	0	0.2	0.1	0.4	6.26	0.282
Galumna lanceata (Oudemans)	F	0	0.1	0	0	0.2	0.7	6.26	0.282
	W	0	0	0	0	0.1	0.6	11.69	0.039

Species		Research data						Kruskal-Wallis test	
		V. 2011	VII. 2011	X. 2011	V. 2012	VII. 2012	X. 2012	*H*	*p*
Gymnodamaeus bicostatus (C.L. Koch)	F	0.1	0	0	0.3	0.2	0.1	6.52	0.259
	W	0	0	0	1.1	0.8	0.3	9.40	0.094
Lauroppia neerlandica (Oudemans)	F	0	0.1	0	0	0	0	5.00	0.416
Liochthonius sp.	F	0.1^{A}	3.5^{B}	0	0	0.1^{A}	0	24.85	0.001
	W	0	0	0	0.1	0	0	5.00	0.416
Metabelba pulverulenta (C.L. Koch)	F	0.1	0.3	0	0	0.7	0	5.21	0.390
	W	0	0	0	0.1	0.1	0.5	5.00	0.416
Microremus brevipes (Michael)	W	0	0	0	0.1	0.1	0	4.07	0.540
Microzetorchestes emeryi (Coggi)	F	0	0.2	0	0.1	0.1	0	3.11	0.684
	W	0	0	0	0	0	0.10	5.00	0.416
Nanhermannia nanus (Nicolet)	F	0.5	0	0	0	0.1	0	7.31	0.199
Oppiella nova (Oudemans)	F	0.1^{A}	0.8^{A}	0.2^{A}	1.4^{B}	1.8^{B}	0	18.11	0.003
	W	0	0	0	0.1	0.1	0.2	3.11	0.684
Oribatula tibialis (Nicolet)	F	1.1^{A}	2.6^{A}	5.3^{B}	33.1^{C}	8.5^{B}	6.3^{B}	35.52	0.000
	W	0.1^{A}	0	0.2^{A}	10.6^{B}	3.5^{BC}	1.3^{C}	35.78	0.000
Pergalumna nervosa (Berlese)	F	0	0	0	0.1	0	0	5.00	0.416
Phthiracarus longulus (C.L. Koch)	W	0.1	0	0	0	0	0	5.00	0.416
Quadroppia quadricarinata (Michael)	F	0	0	0	0.1	0	0	5.00	0.416
	W	0	0	0	0	0.1	0	5.00	0.416
Rhysotritia duplicata (Grandjean)	F	0.2	0	0	0	0	0	10.17	0.071
Scheloribates laevigatus (C.L. Koch)	W	0	0	0	0.1	0	0	5.00	0.416
Steganacarus carinatus (C.L. Koch)	F	0.1	0	0	0	0.2	0	4.07	0.540
Suctobelba sp.	F	0.4^{A}	5.7^{B}	0.5^{A}	1.5^{A}	1.4^{A}	1.8^{A}	13.26	0.021
	W	0	0.1^{A}	0.1^{A}	0.3^{A}	1.2^{B}	0.6^{A}	10.94	0.053
Tectocepheus velatus (Michael)	F	7.9^{A}	105.2^{B}	3.4^{AC}	4.6^{AC}	1.6^{C}	2.1^{C}	30.76	0.000
	W	0	0.3^{A}	0	4.0^{B}	1.4^{B}	1.3^{B}	24.71	0.000
Trhypochthonius tectorum (Berlese)	F	0.2^{A}	1.0^{A}	0	0	0	0.1^{A}	12.66	0.027
Trichoribates trimaculatus C.L. Koch	F	0.1	0	0	0.1	0	0.1	3.11	0.681
	W	0	0	0	0	0	0.1	5.00	0.416

Explanations: see tab. 1.
Source: own research data

Analysis of occurrence of selected *Oribatida* species

On the microplots covered with litter substrate, a dominant oribatid mite on the first sampling date was *Chamobates schuetzi* – *D*=40%. Its number markedly decreased in the autumn of 2011 and then increased to its peak level of 14.6 individuals per 50 cm^3 (Table 2). In the woods chips, this species was rare in the first year of the study but its density was rising gradually over the second year. It is a common species in Poland, belonging to the *Oribatida* of Scots pine forest (Seniczak and Solhøy 1988).

In the litter substrate in May 2011, the second dominant species was *Tectocepheus velatus* (*N*=7.9 individuals per 50 cm^3, *D*=38%). Over the next two months, its number increased by more than 13 times and then stabilized at 1.6-4.6 individuals per 50 cm^3. It was rare in the pine chips in the first year of the study. Over the second year, its number in this substrate rose markedly, up to 1.3-4.0 individuals per 50 cm^3. *Tectocepheus velatus* is a common soil oribatid mite present in various biotopes (Weigmann and Kratz 1981), particularly popular in Scots pine forests (Klimek 1999). It is a parthenogenetic species with short life cycle, high reproduction rate and great ability to colonize new environments (Gulvik 2007, Skubała and Gulvik 2005).

Another species abundant on the investigated microplots was *Oribatula tibialis*. In the beginning, its number in raw humus substrate was rather low, amounting to 1.1-2.6 individuals per 50 cm^3. A significant increase in *O. tibialis* number was observed i the autumn of 2011, when this species was found to be a dominant oribatid mite (*D*=51%). Further increase in its number was reported in May 2012, when it achieved its peak level of 33.1 individuals per 50 cm^3, and its dominance rate was 67 %. On the two last sampling dates, the density of *O. tibialis* was reduced to 6.3-8.5 individuals per 50 cm^3. This species was less abundant in the pine chips. It was rare in the first year of the study, similarly as the other oribatid mites in this experimental variant. A peak in its density was observed in the spring of 2012 (10.6 individuals per 50 cm^3). However, on the next sampling dates its density decreased, similarly as in the pine litter substrate. *Oribatula tibialis* is classified as eurytopic species (Weigmann 1991, Weigmann and Kratz 1981) preferring forest soils (Rajski 1968). It was the most abundant in the second year of the study in birch nursery mulched with raw humus and irrigated (Klimek *et al.* 2013b).

SUMMARY AND CONCLUSIONS

Mite aggregations, observed during a 2-year study in two different substrates intended for soil regeneration, followed different courses of development. Mite density in the shredded pine litter, intended mainly for soil inoculation with edaphon, was similar in the initial and final period of the study. In the pine wood

chips, intended for mulching degraded soils and providing optimal conditions for mesofauna development, mite density was low in the initial period of the study. Then, it gradually increased up to its highest level in spring 2012, and stabilized at a slightly lower level over the next sampling dates.

Oribatid mites were the most abundant mites in the analyzed samples. Mean number of their species *s* in the pine forest litter was comparable at the beginning and at the end of the study cycle, with insignificant differences between individual sampling dates. Different situation was observed for pine chips, where mite species diversity increased over the course of the study and the differences between sampling dates in the first and second year were significant. Oribatid mites frequently observed in both substrates included *Chamobates schuetzi*, *Oribatula tibialis* and *Tectocepheus velatus*.

This study demonstrated that forest raw humus, double-shredded with a garden shredder, is a good inoculum for inoculation of degraded soils with edaphon. Fresh pine chips are gradually colonized by the mesofauna, the first identified species include predatory *Mesostigmata*, and *Oribatida* are the most abundant in the second year.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the considerable help provided by the Forest Nursery Białe Błota and the Forest Inspectorate in Bydgoszcz.

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Received: 04.01.2015
Accepted: 14.05.2015