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Effects of immission load on spiders living on black pine

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Abstract. The effects of immission load on spiders living on black pine (*Pinus nigra* Arnold) were studied in a town in Western Hungary. In three sampling periods, trunk-trap catches of spiders from black pine trees planted in a control urban area were compared with those from black pines planted in an area with high immission load. In the first two sampling periods – when apart from the exclusive dwellers on or under bark of the black pine, foliage-dwelling spiders overwintering under the bark (facultative bark-dwellers) could also be trapped – the species richness and the Shannon diversity were significantly higher in the control area than in the area highly affected by immission. In these sampling periods, the composition of spider assemblages also significantly differed in the studied areas. In the third sampling period – when only the exclusive bark-dwellers could be trapped because facultative bark-dwelling spiders returned to the foliage after the overwintering – the species richness, Shannon diversity and the composition of spider assemblages were similar in the differently affected areas. Our results suggest that spiders living exclusively on or under the bark are not significantly influenced by immission load, while foliage-dwellers are sensitive to immission. Indicator species analysis also proved the clear classification of the responses of spiders to immission load. The probable direct and indirect effects of immission, causing differences in the species richness, diversity and the composition of spider assemblages, are also discussed.

Key words: bark, black pine (*Pinus nigra*), diversity, immission, species richness, spiders

Introduction

With the exponential increase of human activities and technical progress, many procedures and equipment are developed that alter and damage the natural environment. Intense industrial production and traffic increasing quantities of sulphur dioxide (SO_2), nitrogen oxide (NO_x), cyclic hydrocarbons and heavy metals (e.g. lead, zinc, cadmium, etc.) in the air also threaten species directly and indirectly through destruction and change in their habitats. In this respect, primarily the towns are regarded as especially dangerous areas.

The town called Szombathely, located in West-Hungary, is also affected by gas (SO_2, NO_x) and dust (including heavy metals) pollutants emitted by local events (heat and power generating plants, car traffic, etc.). The goal of our study was an analysis of the spider assemblages in two lines of black pine trees planted in areas differently affected by immission. Spiders are favourable subjects for comparative ecological studies, because they are diverse and abundant, and their ecology and systematics are relatively well-known. Moreover, spiders can be found almost everywhere, and being mobile and relatively short-lived, they may adjust rapidly to changes in abiotic and biotic environmental factors. Since they are predaceous, there is the potential for biological concentration of toxic matter such as air pollutants (e.g. heavy metals). Previous studies have also indicated that spiders are sensitive to air pollution, therefore they are useful as an indicator group to assess human influence on natural habitats (Clausen 1986; Szinetár 1993). Our paper was also motivated by the fact that there have been relatively few investigations concerning the effect of immission load on arthropods, especially on spiders (Freitag and Hastings 1973; André 1977; André et al. 1982; Clausen 1984b).

In this paper, we have presented a comparative ecological analysis of spiders living on black pine in areas differently affected by immission. We examined whether there were any differences in structure, species richness or diversity of spider assemblages in scarcely and highly affected areas. Furthermore, we have also analysed the responses of spiders living on different parts of the pine concerning immission load.

Materials and methods

Study area and sampling

The sampling sites were located in the town of Szombathely in West-Hungary (see Horváth and Szinetár 1998). Two lines of planted black pine (*Pinus nigra* Arnold) trees were selected for the analysis, according to the extent of immission load (Table 1):

1. Control urban area: Gagarin Avenue. This avenue is surrounded by free-standing houses. There is only a small road near it, therefore the immission load is low.
2. Highly affected area: Paragvári Street. This sampling site is situated next to a main road.

Due to heavy traffic, the immission load is high in this part of the town. The difference in the amount of SO_2 and dust was not significant between the two areas, either in a heating period or in a period without heating. The amount of NO_x appears to be the crucial factor regarding the immission load of the localities (Table 1).

Samples were taken with trunk-traps made of waved cardboard. Not only the exclusive dwellers on or under the bark of black pine, but also the foliage-dwelling spiders can be sampled and studied by using trunk-traps, because foliage-dwelling spiders overwinter under bark (Wunderlich 1982). Traps were 15 cm wide and placed 2 m above the ground on the trunk. Belts were placed around the trunk, their waved surfaces facing the trunk and covering the whole outline of the tree, as if a kind of

Table 1. Mean quantities of the air pollutant in areas with different immission loads in the studied town. The heating period covered the months from November to March, while the period without heating includes the months from April to June.

	Heating period in 1995–1996			Period without heating in 1996		
	SO_2 (μg m^{-3})	NO_x (μg m^{-3})	Dust (g m^{-2} per 30 days)	SO_2 (μg m^{-3})	NO_x (μg m^{-3})	Dust (g m^{-2} per 30 days)
Control area with low immission load	10.45	37.07	2.41	1.90	16.77	6.11
Area with high immission load	13.05	46.64	2.75	2.44	31.71	6.23

artificial bark (Bogya 1995). In October 1995, 30 trunk-traps were placed in each sampling site on different trees. Traps were checked three times, in January, April and June of 1996, in order to study the seasonal activities of spiders. In each period 10 samples were collected from each site. Trapped spiders were identified by species using standard keys (Loksa 1969, 1972; Heimer and Nentwig 1991; Roberts 1995). Some juveniles were kept at 20–22 °C and fed fruit-flies (*Drosophila melanogaster*) until they completely developed and we could identify them. Concerning the species, the nomenclature of Platnick (1997) was followed.

Data analyses

To test effects of immission load on spiders living on black pine, the species richness and Shannon diversity (e.g. Pielou 1975) was analysed per trap. The unpaired t-test was used to detect differences in the spider species richness and the diversity per trap, between the two sampling sites in the three sampling periods (Sokal and Rohlf 1981).

Non-metric multidimensional scaling (NMDS) (Gordon 1981) using the Bray–Curtis index of percentage similarity (Ludwig and Reynolds 1988) was used for abundances to assess similarities in spider assemblages among the traps, in the three sampling periods.

To find indicator species and species assemblages characterising the sites with different immission load, the IndVal (Indicator Value) approach was used (Dufrêne and Legendre 1997). This method derives indicator species and species assemblages from hierarchical or non-hierarchical site classification. We obtained the site classification from the NMDS using the Bray–Curtis similarity for abundances. Based on this site classification, IndVal identifies the indicator species corresponding to the various groups. Indicator species are defined as the most characteristic species of each group, found mostly in a single group of the classification and present in the majority of the sampling points belonging to that group. The method is robust to differences in the absolute abundances of different taxa. Furthermore, by using a

Monte Carlo permutation test, it is possible to assess the statistical significance of the species indicator values (Legendre and Legendre 1998; McGeoch and Chown 1998).

In the three sampling periods samples were analysed separately, because in each period the 10 traps were collected from different black pines in both habitats. Therefore, the pooling of samples for the entire sampling period is not acceptable in respect to data analysis.

Results

During the study, all together, 1382 individuals belonging to 26 species and to another 12 taxa determined on the level of genera were trapped (Table 2).

Species richness of spiders was significantly higher in the control sampling site with low immission load (Gagarin Avenue) than in the highly affected site (Paragvári Street) during the first two sampling periods (t = 5.4187, df = 1, 18, $P < 0.0001$; t = 2.2361, df = 1, 18, $P = 0.0383$, respectively; see Figures 1A,B), while in the third period, the difference in species richness between the sampling sites was not significant (t = 0.2182, df = 1, 18, $P = 0.8297$; Figure 1C).

The same basic pattern was obtained regarding the Shannon diversity. Spider diversity was significantly higher on Gagarin Avenue (control area with low affection) than on Paragvári Street (high immission load) during the first two sampling periods (t = 4.1691, df = 1, 18, $P = 0.0006$; t = 3.6631, df = 1, 18, $P = 0.0018$, respectively; see Figures 1A,B), while in the third period the difference in diversity between the above two sites was not significant (t = 0.0483, df = 1, 18, $P = 0.9620$; Figure 1C).

The results of the ordination (NMDS) show that in the first two sampling periods the spider assemblages of areas with low and high immission load separated from each other. These facts suggest that sampling sites with a different extent of immission load have different species assemblages (Figures 2A,B). In the third sampling period, however, the spider assemblages of differently affected areas were similar to each other (Figure 2C).

The collected spider species can be divided into three groups by characterisation of habitats by indicator species (Table 3A–C): (1) habitat generalists that existed numerously in all habitats, suggesting that they are not sensitive to immission load (e.g. *Philodromus* (*aureolus*) species group, *Nuctenea umbratica* (Clerck 1757); *Clubiona genevensis* L. Koch 1866; *Theridion mystaceum* L. Koch 1870; *Scotophaeus scutulatus* L. Koch 1866), (2) species that are sensitive to immission were recorded exclusively, or were the most abundant in the control sampling site with low immission load (e.g. *Anyphaena accentuata* (Walckenaer 1802); *C. pallidula* (Clerck 1757); *Cheiracanthium mildei* L. Koch 1864) and (3) in the second sampling period, one species (*T. tinctum* [Walckenaer 1802]) seems to have preferred the area with high immission load, although its indicator value was not significant.

Table 2. Number of individual spiders (female/male [juvenile]) collected in the differently affected areas.

Species	Control area with low immission load	Area with high immission load	∑
Segestriidae			
Segestria bavarica C.L. Koch, 1843	2/2(1)	0/0(0)	2/2(1)
Dysderidae			
Harpactea rubicunda (C.L. Koch, 1838)	4/1(0)	0/0(0)	4/1(0)
Harpactea sp.	0/0(1)	0/0(0)	0/0(1)
Theridiidae			
Anelosimus vittatus (C.L. Koch, 1836)	0/0(3)	0/0(0)	0/0(3)
Dipoena melanogaster (C.L. Koch, 1837)	0/0(0)	0/0(1)	0/0(1)
Dipoena sp.	0/0(2)	0/0(0)	0/0(2)
Steatoda bipunctata (Linnaeus, 1758)	23/9(1)	7/2(3)	30/11(4)
Theridion blackwalli O.P.-Cambridge, 1871	3/0(2)	1/0(5)	4/0(7)
T. impressum L. Koch, 1881	0/0(0)	0/0(1)	0/0(1)
T. mystaceum L. Koch, 1870	0/0(20)	0/0(79)	0/0(99)
Theridion sp.	0/0(1)	0/0(0)	0/0(1)
T. tinctum (Walckenaer, 1802)	0/0(88)	4/0(107)	4/0(195)
Linyphiidae			
Linyphiidae sp.	0/0(1)	0/0(0)	0/0(1)
Moebelia penicillata (Westring, 1951)	3/1(0)	0/0(0)	3/1(0)
Tetragnathidae			
Tetragnatha sp.	0/0/(0)	0/0(1)	0/0(1)
Zygiella sp.	0/0(1)	0/0(1)	0/0(2)
Araneidae			
Araneus sp.	0/0(0)	0/0(1)	0/0(1)
Gibbaranea sp.	0/0(8)	0/0(2)	0/0(10)
Nuctenea umbratica (Clerck, 1757)	2/0(8)	5/1(6)	7/1(14)
Agelenidae			
Tegenaria ferruginea (Panzer, 1804)	0/0(2)	0/0(0)	0/0(2)
Dictynidae			
Lathys humilis (Blackwall, 1855)	1/0(3)	0/0(3)	1/0(6)
Nigma walckenaeri (Roewer, 1951)	2/0(0)	0/0(0)	2/0(0)
Anyphaenidae			
Anyphaena accentuata (Walckenaer, 1802)	0/0(167)	0/0(0)	0/0(167)
Clubionidae			
Cheiracanthium mildei L. Koch, 1864	1/3(34)	0/0(0)	1/3(34)
Clubiona brevipes Blackwall, 1841	0/0(1)	0/0(0)	0/0(1)
C. genevensis L. Koch, 1866	1/0(23)	1/0(128)	2/0(151)
C. pallidula (Clerck, 1757)	0/0(362)	0/0(0)	0/0(362)
Clubiona sp.	0/0(0)	0/0(1)	0/0(1)
Gnaphosidae			
Micaria subopaca (Westring, 1862)	1/0(0)	0/0(3)	1/0(3)
Scotophaeus scutulatus (L. Koch, 1866)	2/1(37)	0/0(37)	2/1(74)
Scotophaeus sp.	0/0(1)	0/0(0)	0/0(1)
Philodromidae			
Philodromus dispar (Walckenaer, 1826)	0/0(1)	0/0(0)	0/0(1)
P. margaritatus (Clerck, 1757)	1/0(4)	0/0(0)	1/0(4)
P. rufus (Walckenaer, 1826)	0/0(1)	0/0(7)	0/0(8)
Philodromus spp. (*aureolus*)	0/0(90)	0/0(35)	0/0(125)
Philodromus spp. (*margaritatus*)	0/0(9)	0/0(1)	0/0(10)

Table 2. Continued.

Species	Control area with low immission load	Area with high immission load	∑
Salticidae			
Macaroeris nidicolens (Walckenaer, 1802)	0/0(3)	0/0(0)	0/0(3)
Pseudicius encarpatus (Walckenaer, 1802)	0/0(1)	0/0(0)	0/0(1)
∑	46/17(876)	18/3(422)	64/20(1298)

Discussion

Previous studies on effects of immission have shown that composition, species richness and diversity of spiders are related to the extent of immission load. Gunnarsson (1988) pointed out that pollution has an impact on the composition of spider assemblages. Clausen (1984b), studying the number of spider species, also reported that there is a very strong, significant negative correlation between the species richness of spiders and the SO_2 concentration in the air; e.g. when increasing the SO_2 level, the number of spider species decreases. However, Gilbert (1971) did not find that the species richness of spiders significantly correlated to SO_2 concentration, but the number of individuals as well as species was surprisingly small, although he applied only visual detection as a 'sampling method'. Apart from SO_2 concentration, spiders were studied in relation to the level of heavy metals. Effects of heavy metals on spider assemblages were mentioned only in cases of extremely high pollutant concentration (Strojan 1978; Bengtsson and Rundgren 1984; Clausen 1984b). Clausen (1984b, 1986), analysing the impact of immission on spiders, showed that there are no significant relationships between the Shannon and Simpson diversity and the extent of immission load. Therefore, he argued that diversity indices should be used with caution, if used at all in studies analysing effect of immission load. Clausen (1986) reasoned that results of Lebrun (1976) and André (1977) are contradictory; they found a positive and a negative correlation, respectively, between the Shannon diversity and the pollution level. However, our results proved that immission load has a significant effect on the composition, species richness and the diversity of spider assemblages (Figures 1 and 2).

Arthropods living in vegetation in polluted areas may be affected directly and/or indirectly by immission load. Direct effects of pollutants comprise their toxic influences on animals, whereas indirect effects may be a result of changed habitat structure and environmental conditions. Both types of effects are described in terrestrial arthropod populations (Alstad et al. 1982). Earlier studies (Williamson and Evans 1972; Price et al. 1974; Wade et al. 1980; Bengtsson and Rundgren 1984; Clausen 1984a) noted that heavy metal concentrations in spiders have elevated values in areas highly affected by immission and may cause toxic effects on spiders. Heavy metals in

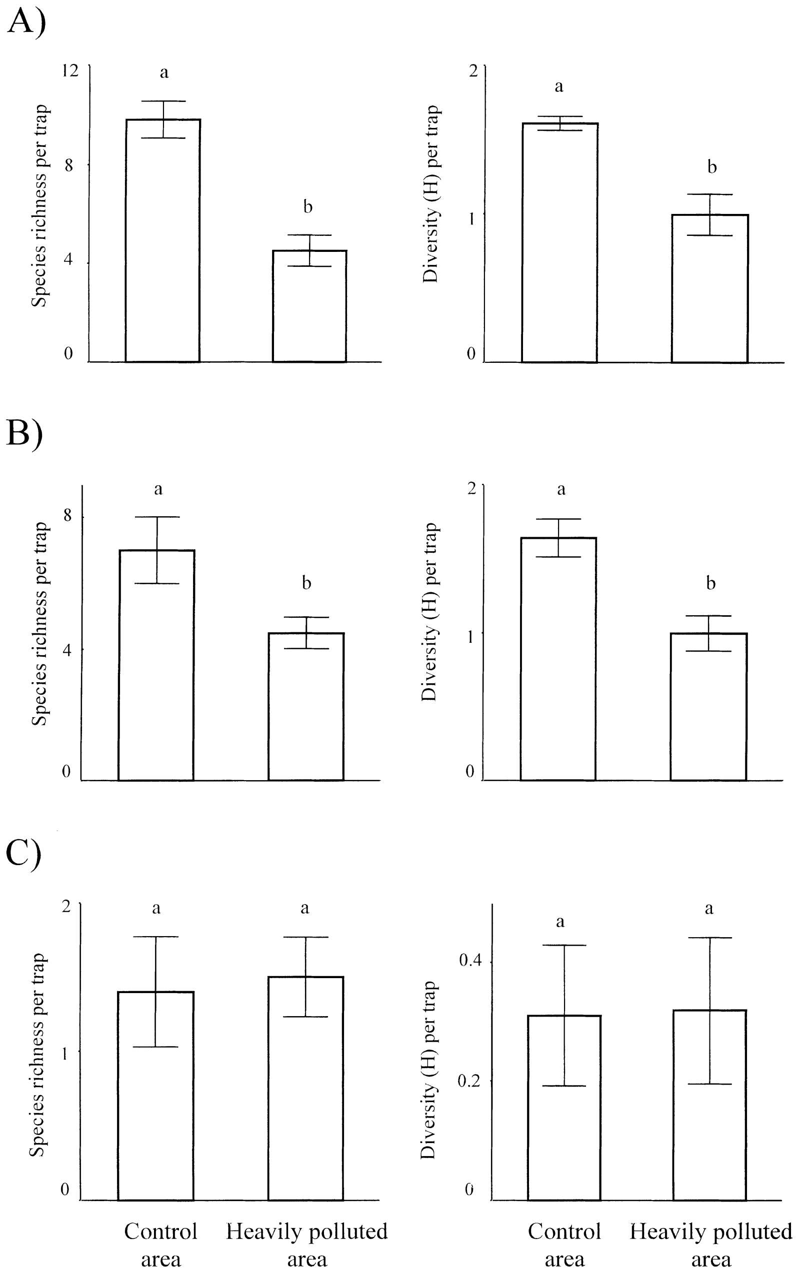

Figure 1. Species richness and Shannon diversity of spiders per trap in the differently affected areas in the first (A), second (B) and third (C) sampling period. Means with different letters indicate a significant ($P < 0.05$) difference by unpaired t-test.

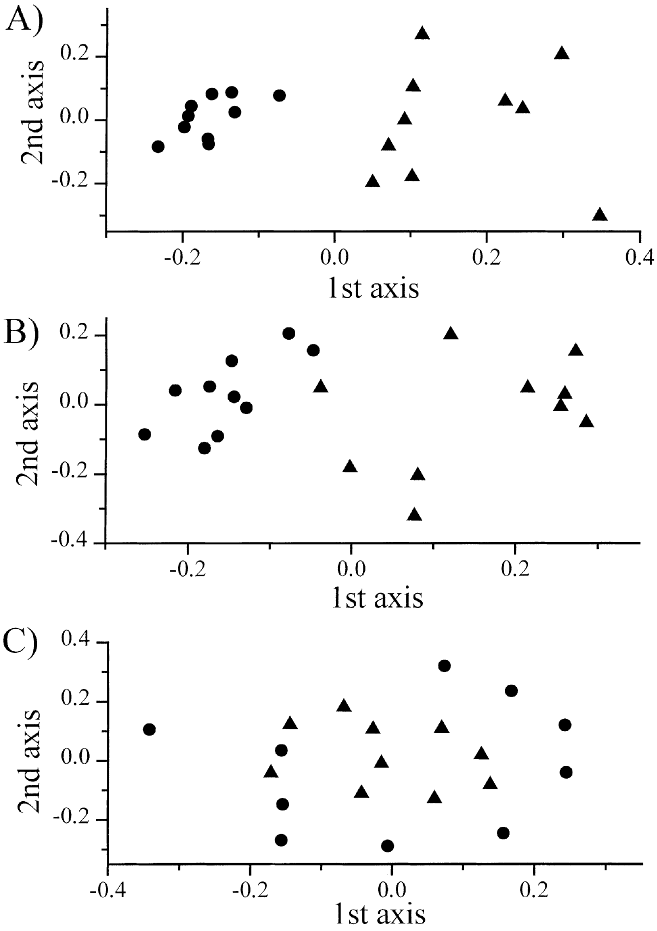

Figure 2. Ordination plot (NMDS by Bray–Curtis index of percentage similarity) of the traps of the differently affected areas in the first (A), second (B) and third (C) sampling period. (•: Control area with low immission load, and ▲: Area with high immission load).

spiders originate directly from the air and from particles deposited on the surface, but indirectly they may arise from consumed prey as well (Clausen 1986). Acid rain may be regarded as a direct effect of pollution on spiders. Gunnarsson and Johnsson (1989), analysing the effects of acid rain on the growth rate of spruce-living spiders in laboratory experiments, pointed out that significant growth reduction occurred only at high acid pH value, so there is weak evidence for the influence of acid rain on spiders. The indirect effects of immission load causing changes in microhabitat structure and environmental conditions probably have a more important influence on structure, species richness and diversity of spider assemblages than direct toxic effects (Clausen 1986; Gunnarsson 1988). Pollution is presumed to cause needle-loss in spruce in Nordic Europe (Andersson 1986), with a consequent change in habitat

Table 3. Two-way indicator table showing the species indicator power of the habitat clustering hierarchy for the species that were represented by more than ten individuals in the first (A), second (B) and third (C) sampling period.[a]

	IndVal		Control area with low immission load	Area with high immission load
(A) Control area with low immission load				
Anyphaena accentuata	100.00	*	153/10	0/0
Clubiona pallidula	100.00	*	320/10	0/0
Cheiracanthium mildei	90.00	*	24/9	0/0
Steatoda bipunctata	88.46	*	23/10	3/3
Both areas				
Philodromus spp. (*aureolus*)	95.00	ns	81/10	32/9
Theridion tinctum	90.00	ns	84/10	87/8
Nuctenea umbratica	55.00	ns	6/6	5/5
(B) Control area with low immission load				
Clubiona pallidula	70.00	*	40/7	0/0
Anyphaena accentuata	50.00	*	14/5	0/0
Cheiracanthium mildei	50.00	*	14/5	0/0
Both areas				
Clubiona genevensis	95.00	ns	23/9	119/10
Theridion mystaceum	85.00	ns	15/7	78/10
Steatoda bipunctata	45.00	ns	10/5	9/4
Philodromus spp. (*aureolus*)	40.00	ns	9/5	3/3
(C) Area with high immission load				
Theridion tinctum	42.31	ns	4/2	22/5
Both areas				
Scotophaeus scutulatus	70.00	ns	38/7	34/7

[a] In the row for each species, the first number indicates the number of specimens present and the second number corresponds to the number of traps where the species is present, in this sample group. The IndVal column indicates the species indicator value for the corresponding clustering level, which is the maximum indicator value observed in all the clustering hierarchy. ns: not significant; * $P < 0.05$.

structure for spruce-living spiders and other arthropods. Gunnarsson (1988) studying the composition of spiders in two close spruce stands, having relatively high and low percentages of needle-loss, respectively, demonstrated that needle-loss in spruce may have an impact on the composition of spider assemblages. He revealed that raptorial spiders are associated with needle-dense branches, since these branches offer good hiding-places for the spiders between the needles. Another explanation for the differences in spider composition may be bird predation, which is known to cause high mortality among spruce-living spiders (Askenmo et al. 1977; Gunnarsson 1983). In the stand with high percentages of needle-loss, spiders might be more vulnerable to bird predation, since they should be easier to detect on branches with few needles (Gunnarsson 1988). An alternative explanation for the differences in spider composition is the microclimate, which may be favourable (wetter and colder) for spiders in the sheltered stand, having low percentages of needle-loss, than in the exposed stand (high percentages of needle-loss) (Gunnarsson 1988). Although we did not study the direct and indirect effects of immission load, presumably the above discussed impacts are also significant in Central Europe.

Our results seem to be in accordance with several previous findings, which have shown that the indirect effects of immission load (changes in habitat structure and environmental variables) are the most important determinants in the composition, species richness and diversity of spider assemblages. In the third sampling period, we detected that the composition, species richness and the diversity of spider assemblages in the control and highly affected areas were not significantly different (Figures 1C and 2C). This can be explained by the fact that in this period the most abundant spiders were those living exclusively on or under the bark of the black pine (e.g. *S. scutulatus* L. Koch 1866), in both areas. Since, in this period, foliage-dwelling spiders that use the bark only for overwintering (facultative bark-dwellers) returned to the foliage (see Table 3C). These results suggest that spiders living exclusively on or under the bark are not significantly influenced by immission load. The bark is presumably a suitable hiding-place, therefore the exclusively bark-dwelling spiders are only moderately exposed to the direct effects of immission. Moreover, the influences of immission load on the microhabitat structure and microclimate (indirect effects) of the bark may not be considerable. On the other hand, in addition to the exclusive bark-dwelling spiders, foliage-dwellers were also sampled in the first two sampling periods. In these periods, differences in the composition, species richness and diversity of the spider assemblages between the two studied black pine stands were significant (Figures 1A,B and 2A,B). These findings can be explained by the fact that some species (e.g. *A. accentuata* (Walckenaer 1802); *C. pallidula* (Clerck 1757); *C. mildei* L. Koch 1864) completely disappeared or their abundance considerably decreased in the highly affected area (Table 3). The mentioned species are facultative bark-dwelling, so it is presumable that the above-discussed direct and rather indirect effects of immission load influenced their distribution.

The results of the indicator species analysis proved a clear classification of the responses of spiders to immission (Table 3). In the first sampling period, *Theridion tinctum* (Walckenaer 1802) was identified as habitat generalist (Table 3A), while in the second period it seemed to prefer the area with high immission load (Table 3B). This contradiction may derive from biased sampling. In our opinion, this foliage-dwelling species is probably a habitat generalist. However, in the black pines located in the control area (Gagarin Avenue), the individuals of *T. tinctum* (Walckenaer 1802) may have returned earlier to the foliage after the overwintering, and this return contributed to the decrease of its abundance in this area. Our assumption was verified by the fact that the indicator value of this species was statistically insignificant (Table 3B), and that is its presence cannot be unambiguously predicted in the area with high immission load. An analogous case can be observed concerning the species *Steatoda bipunctata* (Linnaeus 1758). In the first sampling period it was a significant indicator species in the control area (Table 3A), while in the second period it was regarded as a habitat generalist (Table 3B). *S. bipunctata* (Linnaeus 1758) is probably a foliage-dwelling species that is sensitive to immission load, but in the area with low immission load (control area) its individuals also returned to the foliage earlier following the overwintering, which resulted in a decrease of its abundance. Earlier studies (Luczak 1984; Bengtsson and Rundgren 1984; Clausen 1984b) indicated that spider species of families Linyphiidae and Clubionidae are more common in areas with high immission load. Regarding species of the family Linyphiidae, our results could not confirm this finding because of the limited number of individuals in the samples (Tables 2 and 3). However, concerning the species of the Clubionidae family, we obtained an opposite result, since certain species of this family (*C. mildei* L. Koch 1864; *C. pallidula* (Clerck 1757)) responded the most sensitively to the immission load (see Tables 2 and 3).

Our results proved that spiders are a suitable group for biological monitoring and indication, and for assessment of biodiversity. However, further manipulative and non-manipulative studies are needed to identify factors that are responsible for changes in structure, species richness and diversity of spider assemblages in areas affected by immission. Only with the full knowledge of these facts can we prepare management and conservation plans in the affected areas that stop or reduce environmental damage.

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