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Regular research paper

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FOREST LITTER BACTERIA: RELATIONSHIPS WITH FUNGI, MICROFAUNA, AND LITTER COMPOSITION OVER A WINTER-SPRING PERIOD

ABSTRACT: This paper presents data on temporal and spatial variability and ecological interactions of bacteria in a Scottish woodland over a winter – spring period (January – April). The study sites covered an area of 1 ha and a range of woodland habitats formed by beech (*Fagus sylvatica*), birch (*Betula pendula* × *pubescens*) and oak (*Quercus petraea*), as well as (one site) a clearance site covered with grass (predominantly *Holcus lanatus*). Subsamples of fresh litter were fragmented for 60 s in a domestic food processor and were subsequently used to estimate the abundance of bacteria by counting under a fluorescent microscope. The preparation of bacterial slides involved staining with DTAF following extraction in phosphate buffer. The data on protozoa, fungi and microinvertebrates were available from parallel research and were obtained using standard methods. Numbers of bacteria appeared to be lower in sites dominated by beech. The highest average bacterial abundance (9.07×10^8 cells g⁻¹ dry litter) was registered in January, and then gradually declined till March, when the lowest (7.37×10^8 cells g⁻¹ dry litter) value was found, before rising again in April. The only significant difference revealed by one-way ANOVA was between January and March results. Both date and site effects were found to be significant by two-way ANOVA, but the date × site interaction was not significant. A number of significant relationships were registered by stepwise regression analysis, ANCOVA, and correlation analysis. In stepwise regression

analysis, the most important predictor for bacterial density was litter moisture content (all months but March). Further significant relationships were revealed with the abundance of fungi, nematodes, and microarthropods, and forest litter fractions of moss, needles, beech seeds and birch leaves. ANCOVA confirmed the importance of interactions with litter composition and moisture content, and the abundance of fungi and microarthropods, and revealed a relationship with the abundance of ciliates. Correlation analysis for separate months revealed various relationships with forest litter composition (including positive – with forest litter fractions of oak leaves, grass, roots, birch leaves, and negative ones – with forest litter fractions of ferns and seeds), and the abundance of other microbiota, including positive with *Folsomia candida* (Insecta, Apterygota, Collembola), fungi, plant and microbial feeding nematodes, tardigrades and enchytraeids, positive and negative with ciliates, and negative with predatory nematodes. Most of these relationships, plus a further correlation with the abundance of amoebae, were also revealed for the combined dataset. It should be noted that some of these interactions (e.g. with % grass, % roots, the density of *Folsomia candida*) were only revealed by correlation analysis, and may therefore be judged as less important than relationships registered by all statistical methods applied. The results of this study highlighted the complexity of multivariate interactions of bacteria in forest litter.

KEY WORDS: forest litter, bacteria, protozoa, fungi, microarthropods, nematodes, ecological interactions

1. INTRODUCTION

Forest litter bacteria are especially important for completion of organic matter mineralization (Dilly *et al.* 2001), which often starts from the fungal stage (Ponge 1991). Bacterial contribution to the decomposition process is complementary to that of other microbiota (Kurihara and Kikawa 1986). A number of previous studies have examined bacterial interrelations with the abundance and/or activity of other biota, e.g. fungi (Ohtonen *et al.* 1992, Sidorova and Velikanov 1997, Zhang and Zak 1998, Moller *et al.* 1999, Okoh *et al.* 2000) fungi and forest litter composition (Kshatriya *et al.* 1994), fungi and actinomycetes (Golovchenko and Polyanskaya 1996, Alekhina *et al.* 2001), fungi and nematodes (Mikola and Sulkava 2001), fungi and microarthropods (Okoh *et al.* 1999), fungi and protozoa (Griffiths *et al.* 2001), fungi, algae, testate amoebae and microarthropods (Frouz *et al.* 2001). However, it seems that field research on forest litter bacteria quantitatively assessing the whole complex of ecological interactions with the abundance and/or activity of fungi, protozoa (flagellates, ciliates, amoebae), nematodes, microarthropods, and forest litter composition is not common. Furthermore, ecological interactions of soil and forest litter microbiota (including bacteria) during colder seasons are particularly understudied, partly due to the assumption that the relationships during the colder periods are negligible owing to the subdued biological activity. However, despite a decrease in biological activity, it does not stop completely, and valuable investigations of soil and litter microbiota have previously been carried out in the conditions of winter, subzero temperatures and/or snowcover, including those which are far more challenging than the conditions in the Scottish Borders (Aitchison 1983, Evens 1992, Kennedy 1993, 1999, Block *et al.* 1994, Itoh 1994, Kopeszki and Trockner 1994, Olear and Seastedt 1994, Lavy and Verhoef 1996, Hopkin 1997, Hodkinson *et al.* 1998,

Castrillo *et al.* 2001, Robinson 2001, Ley and Schmidt 2002, Panicker *et al.* 2002, Zettel *et al.* 2002).

This paper presents the results of a field study focusing on ecological interactions of bacteria in forest litter of the Heron Wood Reserve (Peeblesshire, UK – 55°34'N, 3°49'W) over a winter-spring period. The litter fall in a temperate woodland normally happens in the autumn, and is largely complete by the end of December. Therefore, January was the best time to start the sampling, in order to avoid the risk of results being influenced by changes in litter cover. On the other hand, in late April–May the ground (particularly in birch and mixed plots) may become covered with a new growth of herbaceous plants, which may influence the interactions observed. For these reasons the study was carried out over the January till April period.

2. STUDY AREA, MATERIAL AND METHODS

The Heron Wood reserve (Peeblesshire, Scotland) is part of the Dawyck Botanic Garden situated on the Silurian rock system characteristic of the Scottish Borders. It lies on a NW slope of a hill covered with shallow stony soils, which are acidic and almost lime free. Typical soil acidity is pH 3–4 and typical soil organic content is 10–20%. The climate is characterised by low temperatures occurring between November and mid-March (e.g. in December as low as – 18°C). The average daily air temperatures (measured by a small meteorological station situated approximately 400 m from the site) for the investigation period were 4.2, 4.0, 3.4 and 8.2°C for January, February, March and April respectively. The average rainfall for these months was 3.1, 4.4, 2.2 and 2.9 mm respectively. The previous research at the site was related to the ecology of fungi (Krivtsov *et al.* 2003c, Krivtsov *et al.* 2004a), protozoa (Krivtsov *et al.* 2003a), nematodes and microarthropods (Bezginova *et al.* 2001, Krivtsov *et al.* 2001a, 2003b, Thompson *et al.* 2001) and overall ecosystem functioning (Krivtsov *et al.* 2002, 2004b, Walker *et al.* 2002).

Within the reserve an area of 1 hectare has been designated for the research. In each

quarter of this site two smaller sites 100 m² in size have been designated, making 8 in total. The sites covered a range of woodland habitats dominated by beech (*Fagus silvatica*) (plot 1), beech with some birch (*Betula pendula* × *pubescens*) (plot 8), birch (plots 5,6), mixed vegetation – beech and birch (plot 2), birch and oak (*Quercus petraea*) (plot 3), birch, oak and beech (saplings) (plot 7), and a clearance covered with grass, predominantly *Holcus lanatus* (plot 4). Composition of the forest litter samples reflected the predominant vegetation. In particular, birch leaves were most abundant at sites 5 and 6, while seeds (the fraction dominated by heavy beech seeds) and beech leaves at site 1, and grass at site 4 respectively. The litter collected was relatively wet, with mean moisture content of 74.4 % fw. Moisture content appeared to have a slight tendency to decrease throughout the research period (Table 1), with January values (mean = 75.7%) being significantly ($P = 0.033$) different from April values (mean = 72.2%).

Monthly sampling of forest floor consisted of 32 samples (8 plots, four replicates from each plot) taken on each sampling occasion from points chosen randomly along the border of each plot. Samples were taken in January, February, March and April. Each sample was collected from an area of approximately 10 × 15 cm using a plastic frame. In the laboratory samples were hand sorted, and the sample composition was assessed by measuring the weight of specific fractions. The samples predominantly consisted of forest litter, and the major fractions included beech leaves, birch leaves, oak leaves, conifer needles, lichens, moss, grass, seeds, wood, roots, and unidentifiable (i.e. owing to the stage of the decomposition) plant fragments. Five grams from each sample were taken for nematodes analysis (see below), whilst the remaining litter was split into three aliquots subsequently used for a) measurements of moisture content, b) analysis of bacteria, fungi and protozoa following shredding, c) analysis of microarthropods.

Moisture content of the litter samples was determined as the weight loss following drying for 48 hr at 80°C. Weight of sample provided an index of the thickness of the litter cover. Subsamples of fresh litter (1/3 of

the sample left after removing 5g for nematodes analysis) were fragmented for 60 s in a domestic food processor (Thompson *et al.* 2001), and were subsequently used to estimate the abundance of bacteria by counting under a fluorescent microscope. The preparation of bacterial slides involved staining with DTAF following extraction in phosphate buffer (Alef and Nannipieri 1995).

For statistical analysis the data on fungi and microbiota already partly published were used (Krivtsov *et al.* 2002, 2003a, b, 2004a). The data on protozoa, fungi and microinvertebrates were obtained using standard methods. Subsamples of litter fragmented for 60 s were used to assess the abundance of fungi and protozoa. Concentration of ergosterol, a biomarker for total live fungal biomass, was assayed by HPLC following sonication (Ruzicka *et al.* 1995). The abundance of protozoa was assessed using the MPN method (Bamforth 1995). Microarthropods were extracted from known quantities of unfragmented forest litter using Tullgren funnels and counted under a light microscope (Southwood 1978). Nematodes, enchytraeids and tardigrades were counted under a light microscope following extraction using Baermann funnels from litter fragmented for 5 seconds (Bezginova *et al.* 2001, Thompson *et al.* 2001). The basic data for the above litter variables (litter composition, litter moisture, abundance of organisms are given in Table 1).

3. RESULTS AND DISCUSSION

Numbers of bacteria appeared to be lower in sites covered by beech. The overall results (Fig. 1) show a gradual decline in numbers from January when they were the highest (9.07×10^8 cells g⁻¹ dry litter) to March when they were the lowest (7.37×10^8 cells g⁻¹ dry litter) before rising again in April. It should be noted, that the only significant difference revealed by one-way ANOVA was between January and March results. However, both date and site effects were found to be significant by two-way ANOVA, although the date × site interaction was not significant (Fig. 1).

The data presented here show high numbers of bacteria which is in line with data from other studies. For instance, in a study

Table 1. Data for forest litter variables (mean values and SE for 8 plots) used in statistical analysis. The summary presented here includes also the information on soil nematodes, protozoans and other mesofauna from Krivtsov *et al.* (2001a, b, 2002, 2003a, b). Presence/absence is given on the scale between 0 and 1, e.g. the value 0.5 for flies in February means that the flies then were present in 50% of samples.

Units	Variables	January		February		March		April		January–April	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
g	Sample weight	103.2	5.9	99.7	5.5	117.8	9.9	107.5	8.0	107.0	3.8
% of sample fresh weight	Moisture content	76	1	75	<1	75	1	72	<1	74	<1
	Beech leaves	7.2	1.3	7.3	1.1	8.7	1.7	7.1	1.2	7.6	0.7
	Wood	8.5	1.4	14.6	1.9	12.6	2.6	14.9	2.1	12.6	1.1
	Seeds	10.7	2.7	11.7	2.7	11.9	2.6	10.9	2.2	11.3	1.3
	Roots	11.2	3.8	6.6	2.2	7.1	1.9	5.7	1.7	7.7	1.3
	Moss	8.0	1.9	5.7	1.5	6.4	1.6	4.4	0.9	6.1	0.8
	Cones	0.0	0.0	0.0	0.0	0.0	0.0	0.1	0.1	0.0	0.0
	Lichens	0.0	0.0	0.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0
% of litter fresh weight	Oak leaves	2.2	0.4	4.0	1.1	2.0	0.5	1.8	0.3	2.5	0.3
	Grass	2.4	0.5	2.6	0.8	2.3	0.6	4.0	1.0	2.8	0.4
	Birch leaves	4.2	0.8	4.2	0.7	4.4	0.9	2.7	0.5	3.9	0.4
	Needles	0.0	0.0	0.2	0.1	0.1	0.1	0.1	0.1	0.1	0.0
	Unidentifiable plant fragments	45.0	2.9	40.3	2.2	42.9	2.3	47.4	2.3	43.9	1.2
	Fern	0.0	0.0	0.0	0.0	0.1	0.1	0.1	0.0	0.0	0.0
	Microbial feeding nematodes	2524.4	288.4	213.8	21.9	8837.5	988.2	5036.5	274.0	4192.1	394.5
Ind. (individuals) 100 g ⁻¹ dw of litter	Plant feeding nematodes	1267.1	171.8	123.4	22.3	7527.9	974.7	7750.3	503.6	4236.8	421.8
	Predatory nematodes	18.4	7.5	0.0	0.0	44.5	15.0	32.7	9.3	24.0	5.0
	Tardigrades and enchytraeids	90.2	15.2	15.7	5.7	735.7	95.7	442.8	87.5	326.7	42.1

Units	Variables	January		February		March		April		January–April	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Ind. g ⁻¹ dw	Flagellates	95153.1	32484.9	73309.4	11132.5	54988.6	14943.7	17475.5	1657.2	60231.6	9599.2
	Ciliates	1010.6	229.7	144.0	32.9	38.6	6.6	28.3	3.4	305.4	67.9
	Amoebae	490.9	106.1	1260.3	229.0	36.9	3.9	204.4	17.2	498.1	75.1
Ind. 100 g ⁻¹ dw	<i>Folsomia candida</i>	563.0	95.9	362.1	48.3	229.6	41.3	173.0	27.4	331.9	32.1
	<i>Folsomia quadrioculata</i>	18.5	7.4	11.7	5.7	22.6	8.5	6.8	4.5	14.9	3.4
	Other Collembola	0.2	0.2	1.3	0.8	0.3	0.3	1.0	0.7	0.7	0.3
	Cryptostigmatic mites	180.3	25.1	47.8	7.8	49.7	9.7	54.0	10.2	83.0	8.9
	Mesostigmatic mites	73.3	12.1	23.0	5.1	31.1	4.2	29.3	5.8	39.2	4.1
	Other mites	179.7	28.9	198.3	32.4	202.1	36.1	151.8	25.3	183.0	15.4
	Microarthropod larvae	47.0	8.9	58.0	10.3	43.7	8.8	44.0	6.9	48.2	4.4
µg g ⁻¹ dw	Leaf litter Ergosterol (indicator of fungal biomass)	125.9	9.6	127.1	8.1	96.1	10.6	132.8	6.7	120.4	4.6
Presence/absence	Flies	0.5	0.1	0.8	0.1	0.2	0.1	0.3	0.1	0.5	0.0
Presence/absence	Other insects	0.7	0.1	0.6	0.1	0.6	0.1	0.6	0.1	0.6	0.0

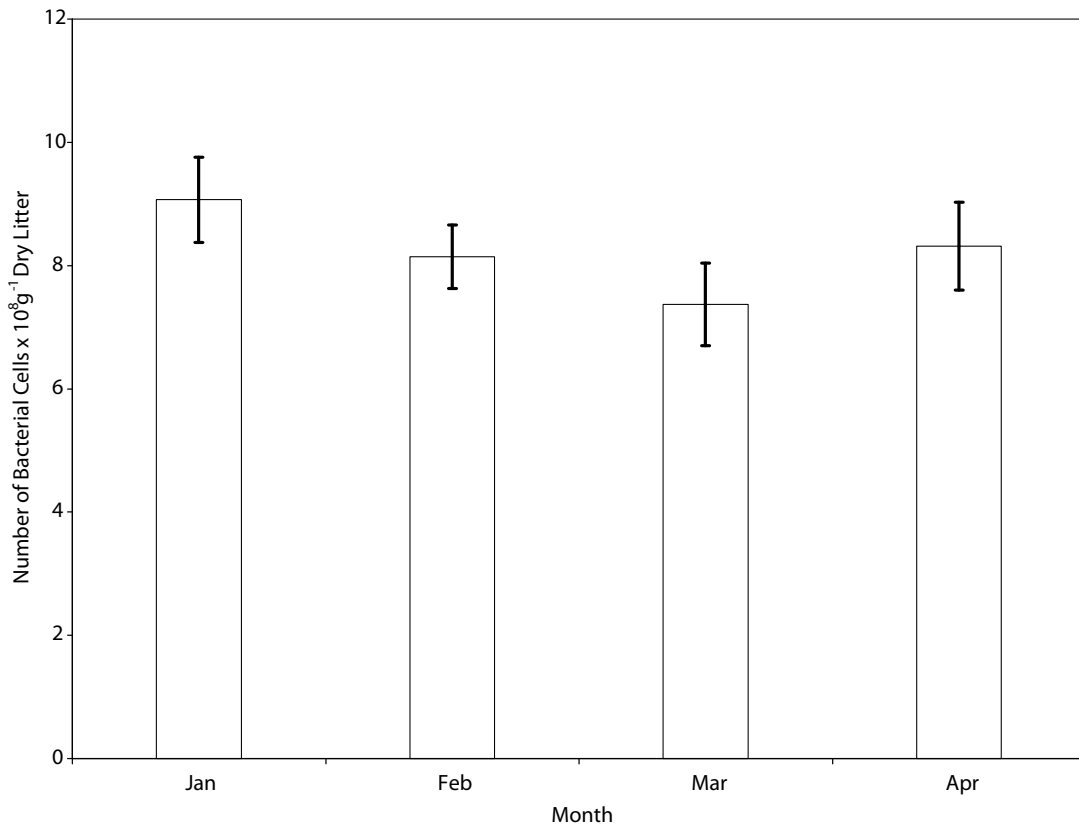


Fig. 1. Temporal changes in mean (for 8 plots) bacterial numbers in forest litter.

of an alpine ecosystem, Lipson *et al.* (1999) found that soil bacteria ranged from 10^8 to 10^9 cells per g dwt, and were higher during the winter (i.e. in comparison with summer values) under the snow cover. Samples of the upper 1–2 cm of arctic soil were reported (Brown *et al.* 1980) to have $4.6\text{--}8.3 \times 10^9$ bacteria per g dwt. High bacterial numbers were also obtained in the experiments conducted with the materials from the upper layers of the soil profile sampled at the Central Forest Reserve in Russia (Alekhina *et al.* 2001).

In our study, bacteria showed minimum values in March (Fig. 1). It should be noted that both temporal and spatial variation in bacterial numbers appeared to be limited (Fig. 1). However, one-way ANOVA revealed a significant difference between January and March results, whilst both date and site effects were found to be significant by two-way ANOVA, suggesting that ecological interactions of forest litter bacteria have been influenced both by habitat characteristics and

by the month of sampling. Previous research (see, e.g. Lipson *et al.* 1999, and references therein) also showed a sudden drop in bacterial numbers just before the start of the plants growing season.

In stepwise regression analysis the most important predictor for bacteria was moisture content of forest litter (Table 2). Importance of the moisture content for microbiota is well known (Rutherford and Juma 1992). Fungi were also among important predictors describing bacterial abundance indicating that both decomposer groups were influenced by the same factors, presumably relating to the weather and resource quality. Bacteria also appeared to be negatively dependent on the abundance of predatory nematodes, mesostigmatic mites and other microarthropods, (Table 2) thus indicating that bacterial numbers might have been controlled by microinvertebrates through digestion of detritus (Kurihara and Kikkawa 1986). It should be noted, however, that both predatory nematodes and mesostigmatic

mites include the groups which prey on the detritivorous groups. Therefore, the presence of these groups might have a positive effect on the abundance of bacteria. Furthermore, the digestion of detritus by microarthropods might lead to the enhancement of bacterial activity, and may not always lead to a decrease in bacterial abundance (Griffiths and Bardgett 1997). The negative relationships might have been indirect, resulting from the habitat preferences of microinvertebrates. Our previous research carried out at the site studied indicated that the abundance of bacteria and the moisture content were the most important variables with regard to the

structure of the soil mesofaunal community (Krivtsov *et al.* 2003b).

The results of stepwise regression analysis have also revealed that bacteria were negatively dependent on the forest floor fractions of moss, conifer needles, and positively on birch leaves and beech seeds (Table 2). These relationships might have been indicative of the relative quality of these resources in relation to bacterial decomposition (see, however, ANCOVA results in relation to the percent of birch leaves in forest litter).

Bacteria have previously been shown to be a significant predictor in the stepwise regression models for fungi, thus supporting

Table 2. Summary of correlations, stepwise regressions and ANCOVA analyses for forest litter variables. For all analyses, exact figures are given where a significant result was obtained on the overall data set, whilst relationships found for only part of the research period are indicated by + or – for positive and negative relationships respectively. Sample (n) = 32 for each month, and sample (n) = 128 for the combined (overall) data set. For Spearman correlation analysis, figures represent values of correlation significant at 95% probability level (two tailed significance $P \leq 0.05$). For stepwise regression analysis and ANCOVA, figures represent regression coefficients followed by T-ratios.

	Correlation Analysis	Stepwise Regression Analysis	ANCOVA
Relationships with Microbiota			
Amoebae	0.24		
Ciliates	0.21		0.04; 0.04
<i>Folsomia candida</i>	0.31		
Fungal ergosterol	0.43	1.14 (3.95)	1.08; 0.001
Meostigmatic mites		–	+
Microbial Feeding Nematodes	+		
Other microarthropods		–	
Plant Feeding Nematodes	+		
Predatory Nematodes	–	–	
Total nematodes		+	
Tardigrades and enchytraeides	+		
Relationships with other variables			
Birch leaves	0.26	+	–12.5; 0.02
Ferns	–		
Grass	0.24		
Litter thickness index			–0.88; 0.04
Moisture content	0.66	25.2 (8.7)	29.9; <0.001
Moss		–	
Needles		–120 (–3.21)	
Oak leaves	0.27		
Roots	+		
Site			0.02
Seeds	–0.23	+	

the results presented here (Krivtsov *et al.* 2002). Both fungi and bacteria are important agents of forest litter decomposition (Kurihara and Kikkawa, 1986), and the positive relationship between them may therefore indicate their simultaneous involvement in the decomposition of palatable materials (Table 2).

It should be noted that previous microcosm experiments with beech leaf litter indicated a possibility of antagonistic interactions between fungi and bacteria due to competition for nutrients (Moller *et al.* 1999). However, considering the results of statistical analysis (e.g. the absence of negative interactions), it appears that during the period of this research at the site studied, bacterial abundance in the forest litter was not considerably influenced by competitive interactions with fungi. The relative importance of fungi and bacteria in the overall metabolism of the microbial community and litter decomposition is known to vary with forest type (Elliott *et al.* 1993), and with a stage of decomposition (Dilly *et al.* 2001). For example, fungi and bacteria were important at different stages of leaf litter decomposition in an alder (*Alnus glutinosa*) forest (Dilly *et al.* 2001). It should be noted that in the present study composite litter samples included materials at various degrees of degradation, which might have masked the real strength of any fungal-bacterial interactions.

It should also be noted that in our parallel investigations bacteria were also found to be a significant predictor for stepwise regression models describing the abundance of microbial feeding nematodes and ciliates, whilst the results of stepwise regression analysis presented in this paper have not revealed the reciprocal relationships. This suggests that the relationships of bacterial abundance with the abundance of microbial feeding nematodes and ciliates registered in the parallel research might have indicated attraction of predators to their prey, whilst the numbers of these particular predators were insufficient to influence to any great extent the bacterial abundance. We have previously (Krivtsov *et al.* 2003b) shown that non-predatory nematodes appear to prefer a high level of bacteria in the habitat studied. It is also well known

that protozoa are the important consumers of microbial biomass (Clarholm 1981, Hunt *et al.* 1987). It is well documented that bacteria can persist even in the presence of active grazers, and that feeding by protozoa and nematodes results in activity in the observed microbial community (Griffiths and Bardgett 1997, Acea and Alexander 1988, Heynen *et al.* 1988). The persistence of bacteria is explained by their compensatory reproduction (Sambanis and Fredrickson 1988), and by the fact that the predator's population growth is negligible once bacterial numbers fall below a threshold density (Sherr *et al.* 1983). However, relationships between bacteria and their predators are complicated (Schönborn 1992), and it was previously shown that the abundance data may correlate more with such factors as the rate of decomposition and content of organic matter, than between themselves (Stout 1973).

The results of ANCOVA analysis were broadly consistent with the pattern revealed by Stepwise Regression Modelling (Table 2). In particular, ANCOVA confirmed the importance of moisture content in determining bacterial population abundance and revealed that the abundance of ciliates and forest litter fungal ergosterol content were among important predictors for bacterial abundance, while mesostigmatic mites appeared to control bacterial abundance in a part of the research period. However, this relationship might have been indirect, resulting, in part, from habitat preferences. It should also be noted that ANCOVA on the combined data showed negative dependence of bacterial abundance on the thickness of the litter layer and on the percent of birch leaves, which was not registered by the stepwise regressions.

The relationships found by the correlation analysis may be subdivided into the following categories. The first category comprises correlations between bacteria and litter components (reflecting association with particular types of forest litter). The second category comprises positive and negative correlations with biological variables reflecting similarities and differences in the occurrence of bacteria and other biological groups studied (Table 2).

Correlation analysis revealed various relationships with litter composition (including positive – with percentages of oak leaves, grass, roots, birch leaves in the forest floor samples, and negative ones with percentages of ferns and seeds) (Table 2).

Positive values of the Spearman correlation coefficient (e.g. with % of birch leaves) indicate that higher abundance of bacteria corresponded to higher values of the variable in question, whilst lower values – to lower values of this variable. Negative values of the correlation coefficient (e.g. with % of fern fragments) indicate that higher abundance of bacteria corresponded to lower values of the variable in question, and *vice versa* (Table 2). Relationships with specific litter fractions are due to the fact that bacteria are important agents of decomposition (Kurihara and Kikkawa 1986), and their relative contribution varies with forest litter type (Elliott *et al.* 1993), and with a stage of decomposition (Dilly *et al.* 2001).

As regards microfauna, positive relationships were registered with the abundance of amoebae, plant and microbial feeding nematodes, tardigrades and enchytraeid worms, and *Folsomia candida*, while a negative correlation with predatory nematodes was revealed (Table 2).

It should be noted that some relationships (e.g. with percentages of grass and roots, and the abundance of *Folsomia candida*) were only revealed by correlation analysis, and may therefore be judged as, perhaps, less important than relationships registered by a combination of the statistical methods applied. It should also be noted, that although the overall correlation with the abundance of ciliates was positive, for a part of the research period a negative significant relationship was found, perhaps indicating that ciliates were exhibiting some controlling influence upon bacterial numbers (Table 2).

Overall, this field study on forest litter bacteria provided a quantitative assessment of their simultaneous ecological interrelations with fungi, protozoan flagellates, ciliates, amoebae, nematodes, microarthropods, and forest litter composition. Some of the relationships described in this paper (e.g. with moisture content, fungi, ciliates

etc.) are well-known, while some others (e.g. with litter composition) are less well-studied. The added value of this assessment is that it relates to the winter-spring period, when the ecological interactions tend to be less investigated on the assumption that the biological activity during the colder periods is subdued. However, despite a decrease in biological activity, it does not stop completely, and it has previously (Clein and Schimel 1995) been shown that, although C and N mineralisation ceased below 0 degrees C, microbial activity in tundra and taiga soils occurred at temperatures as low as -5°C . Valuable investigations of soil and litter microbiota have previously been carried out in the conditions of winter, subzero temperatures and/or snow, including those which are far more challenging than at the site studied in this research (Aitchison 1983, Evens 1992, Kennedy 1993, 1999, Block *et al.* 1994, Olear and Seastedt 1994, Itoh 1994, Kopeszki and Trockner 1994, Lavy and Verhoef 1996, Hopkin 1997, Hodkinson *et al.* 1998, Castriello *et al.* 2001, Robinson 2001, Ley and Schmidt 2002, Panicker *et al.* 2002, Zettel *et al.* 2002). Furthermore, our previous zoological and fungal research conducted over a winter – spring period at the same site showed a number of interesting relationships exhibited by soil and forest litter fungi and microfauna (Krivtsov *et al.* 2002, Krivtsov *et al.* 2003b). Hence, the results presented in this paper provide a valuable complementary insight to the previous work. The ecological interactions of bacteria registered in this research were indicative of the specific conditions of the study, and may, therefore, prove useful for future reference. Further work at the site should compare the relationships found in winter with the patterns observed during the other seasons, and particularly in summer.

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