Folia Microbiol. **49** (2), 105–111 (2004) **http://www.biomed.cas.cz/mbu/folia/**

Microbial Community Structure Along an Altitude Gradient in Three Different Localities

XIAOJUN MA[a], TUO CHEN[b], GAOSEN ZHANG[a], RUI WANG[a]*

[a]*School of Life Sciences, Lanzhou University, Lanzhou 730 000, China*
fax +86 931 891 2561
e-mail wangrui@lzu.edu.cn

[b]*Cold and Arid Regions Environmental and Engineering Research Institute, Chinese Academy of Sciences, Lanzhou 730 000, China*

Received 24 June 2003
Revised version 19 November 2003

ABSTRACT. The microbial community structure along an altitude gradient was investigated in different localities, in Kalasi lake, Urumqi river and Sangong river, Xingjiang (China). The mean numbers of DAPI (4′,6-diamidino-2-phenylindole)-stained cells were lower in Kalasi lake than that in Urumqi river and Sangong river; these differences were attributed to increasing environmental harshness including lower soil organic carbon and nitrogen content, more acidic pH and lower annual temperature. In each locality, the numbers of bacteria and archaea measured with two fluorescence-labeled 16S rRNA oligonucleotide probes (EUB338 and ARCH915) were higher in a coniferous forest and lower in desert vegetation. A significant and positive relationship was found between microbial and soil organic carbon and total nitrogen along the altitudinal gradient, indicating that plant communities and soil nutrients influence the soil microbial structure. The results show that the microbial population in higher latitudinal site was fewer than lower latitudinal one, soil microorganisms were positively correlated to soil organic carbon and total nitrogen, and plant communities had an obviously impact on soil microbes.

Abbreviations

asl	above sea level	DAPI	4′,6-diamidino-2-phenylindole
C_{org}	soil organic carbon	FISH	fluorescence *in situ* hybridization
N_{tot}	total nitrogen		

Soil microorganisms play a vitally important role in soil biogeochemical cycling, decomposition of plant and animal wastes and pollutants in soil and aquatic environments, and symbiotic relationships with higher organisms (Tiedjie *et al.* 1999; Derry *et al.* 1999; Bogoev *et al.* 2002; Kandeler *et al.* 2002; Veselova *et al.* 2003). Consequently, changes in the structure or function of microbial communities may have a major impact on ecosystem activities (Chapin and Korner 1995; Couteaux *et al.* 1995; Swift *et al.* 1998; Elhottová *et al.* 2002; Wahlström and Danilov 2003). The microbial community structure, microbial diversity and microbial groups in a variety of soils were therefore investigated by various methods and techniques (Tiedjie *et al.* 1999; Marilley and Aragno 1999; Kao *et al.* 2003).

However, our knowledge of microbial diversity is limited, as it has been estimated that less than 0.1–10 % of microorganisms have been identified (Head *et al.* 1998). The recent use of molecular biological techniques now allows us to view microbial diversity and community structure without the need of laboratory cultivation (Amann *et al.* 1995; Muyzer and Smalla 1998; Tiedje *et al.* 1999; Stephan and Kowalchuk 2000). For example, fluorescent *in situ* hybridization (FISH) plays an important role in environmental microbial ecology and allows the detection of previously uncultured microorganisms. Labeled oligonucleotide probes can be designed and used to detect microorganisms at different levels (kingdom-, genus- and species-specific) in a variety of environments including pure cultures, environmental enrichments, biofilms, and soils (Amann *et al.* 1995). However, little is known about the influence of large-scale factors such as latitude, biome type, altitude or climatic patterns on the composition of soil microbial communities (Staddon and Trevors 1998; Zhang *et al*. 2002). Using 16S rDNA sequencing, Marilley and Aragno (1999) found that bacterial communities were quite different close to roots and in the bulk soil, respectively. Staddon and Trevors (1998) reported on a decrease in soil microbial diversity across the latitude transect in Canada (from southern to northern) resulting from a gradient of environmental conditions such as decreasing nutrient con-

*Corresponding author.

tent, less productive plant growth, and more acidic soil pH. Altitudinally defined climatic and soil factors are deemed to be primary determinants of change in species composition and community structure in undisturbed mountains (Whittaker 1975). Increasing altitudebrings about significant environmental gradients in both abiotic (temperature, precipitation, atmospheric composition, *etc*.) and biotic factors (vegetation, biodiversity and composition). Consequently, soil characteristics and soil microbial communities may differ among plant communities along an altitudinal gradient. As the main input of organic matter in terrestrial ecosystems, plants are thought to have a profound affect in steering soil communities and processes (Insam and Domsch 1988; Wardle *et al*. 1998).

In this paper, the microbial community structure of different vegetations was investigated by fluorescence *in situ* hybridization (FISH) staining technique along an altitude gradient in Xinjiang which provides an excellent model for such investigation.

MATERIALS AND METHODS

Study area. The study sites were located in Kalasi Lake (87°04′E, 48°49′N), Urumqi river basin (86°49′E, 43°06′N) and Sangong river basin (88°20′E, 43°40′N) of Xinjiang (China; Fig. 1).

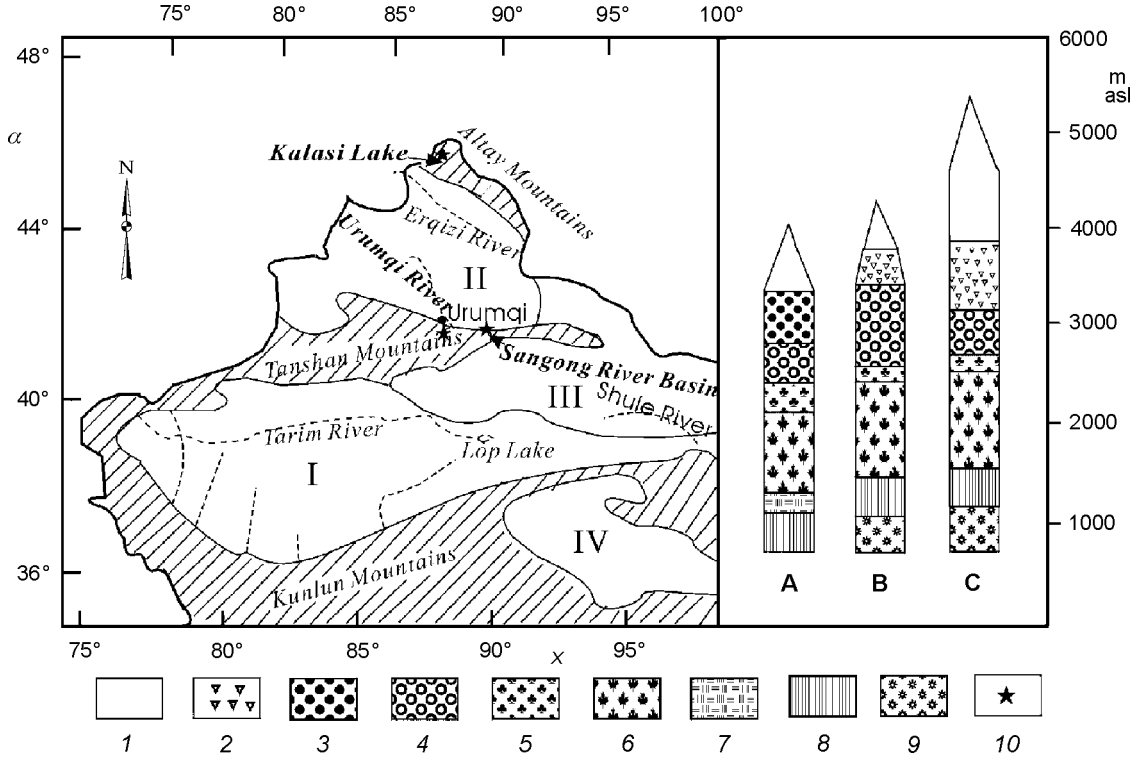

Fig. 1. *Left*: the diagram of study area; desert landscape, surrounded by Altay mountains, Tianshan mountains and Kunlun mountains:
I Tarim basin II Jungar basin
III Xinjiang and Gansu Gobi IV Qaidam basin

Right: vegetation vertical zonation (the location of sampling):
A Kalasi lake B Urumqi river C Sangong river

Bottom: vegetation type (zones):
1 glacial and perennial snow
2 alpine tundra
3 alpine cushion vegetation
4 alpine meadow
5 subalpine meadow
6 montane forest
7 meadow grassland
8 grassland
9 grassland
10 sampling sites

Slopes and soils. The slope gradient is sharp in all three places and mostly ranges from 30 to 60°. The distance from the base altitude (900 m asl) in Sangong river to the top one (3600) is about 80 km. More than 76 % of the total area of the Urumqi river basin is mountains. The soil types and soil properties are changed with altitude. In Kalasi lake, with increasing altitude, the soil types range from montane brown soils (<800 m asl) to montane chestnut soils (800–1200), montane chernozem (1200–1500), montane gray-

wooded soils (1500–2400), to subalpine meadow soils (2400–2800), alpine meadow soils (2800–3000) and finally to alpine ice-bog soils (>3000). Similarly, soil type gradients in the Urumqi river basin within the studied area change from montane chestnut soils (1300–1800 m asl) to montane gray-brown forest soils (1800–2800), subalpine meadow soils (2800–3200), alpine meadow soils (3200–3800) and finally to alpine cold-desert soils (3800–4000). Sangong river basin also has the same altitude-associated sequence of soil types including montane brown soils, montane chestnut soils, montane gray-brown forest soil, sub- and alpine meadow soils.

Vegetation zones. The altitude-associated difference of soil properties and other environmental factors give rise to large variations in vegetation development and dominant plants that are characteristic of vertical zonation (Fig. 1). The dominant plants are listed in Table I.

Table I. Dominant plants and vegetation zonation in Kalasi lake, Urumqi river basin and Sangong river basin of Xinjiang, China

Location	Vegetational zonation[a]	Dominant plants
Kalasi lake	MGB	*Stipa* spp., *Galium verum*, *Potenitla deabata*, *Oxytropsis* sp., *Caragana* spp., *Spiraea hypericifolia*, *Artemisia* spp.
	MFB	*Larix sibirica*, *Picea obovata*, *Abies sibirica*, *Pinus sibirica*, *Betula pendula*, *Lonicera* sp., *Aconitum excelsum*
	SMB	*Betula rotundifolia*, *Salix glauca*, *Poa altaica*, *Alopecurus songoricus*, *Phlomis orecophila*, *Allium semenovii*, *Polygonum nitens*, *Leonthopodium ochroleucum*
	AMB	*Kobresia bellardii*, *K. capillifoila*, *K. capilliformis*, *Carex melanantha*, *C. stenocarpa*, *Festuca* sp., *Polygonum* spp.
	ATB	lichens, *Poa alpina*, *lagotis altaica*, *Polygonum viviparum*, *Gentiana algida*
Urumqi river basin	DGB	*Stipa capillata*, *Artemisia borotaensis*, *A. arenaria*, *Aster altaicus*, *Allium polyrrhizum*, *Carex pachystilis*, *Caragana microphylla*
	MGB	*Stipa capillata*, *Festuca sulcata*, *Poa stepposa*, *Koeleria gracilis*, *Caragana frutex*, *Gallium vertum*, *Potentilla anserine*, *Spiraea hypericifolia*
	MFB	*Picea schrenkiana*, *Betula tianshanica*, *Populus tremula*, *Sorbus tianshanica*, *Salix xerophila*, *Cotoneaster melanocarpa*, *Lonicera* spp., *Poa* spp., lichens
	A&SMB	*Carex atrata*, *C. cobresifolis*, *Kobresia capillifoila*, *K. capilliformis*, *Poa alpine*, *Polygonum viviparum*, *Thalictrum alpinum*, *Festuca supina*
	ACB	*Sibbaldia tetrandra*, *Thylacospermum caespitosum*, *Poa relax*, *Draba alpine*, *Callianthemum alatavicum*, *Potentilla hololeuca*
	ATV	*Kobresia capillifoila*, *Rhodiola algida*, *Aster alpinus*, *Androsae tapete*, *Thylacospermum caespitosum*, *Thalictrum alpinum*
Sangong river basin	MGB	*Stipa capillata*, *Festuca sulcata*, *Iris ruthenica*, *Koeleria gracilis*, *Spiraea hypericifolia*, *Rosa bergeriana*, *Sabina vulgaris*
	MFB	*Picea schrenkiana*, *Betula tianshanica*, *Sorbus tianshanica*, *Salix xerophila*, *Rosa* spp., *Cotoneaster* spp., *Lonicrea spp.*, *Fragaria vesca*, *Rubus saxatilis*, *Poa nemoralis*
	SMB	*Kobresia pamiroalaica*, *Poa alpine*, *festuca supina*, *Alchemilla cyrtopleura*, *Geranium psedosibiricum*, *Phlomis oreophila*
	AMB	*Kobresia capilliformis*, *Carex stenocarpa*, *Polygonum viviparum*, *Leontopodium alpinum*, *Gentiana tianshanica*
	ATV	*Saussurea involucrate*, *Saxifraga hirculus*, *Papaver croceum*, *Primula algida*, *Draba alpina*

[a] A alpine B belt C cushion D desert F forest G grassland M montane S subalpine T tundra V vegetation

Regional climate. Basically, Kalasi Lake belongs to the cold temperate semi-arid continental climate while the other two localities have a temperate desert climate in the foothill. With increasing altitude, the annual mean temperature and mean annual rainfall gradually decrease. Permafrost and glacial area occurr at the top of mountain. The amount of available climatic data from regional meteorological stations are limited. The recorded annual mean temperature is –4.0 °C and the average annual precipitation is 500 mm at Kalasi lake (1300 m asl). Annual mean temperature and rainfall in Sangong river are 4.8 °C and 530 mm (1911 m asl), and 5.9 °C and 187 mm (700 m asl), respectively, while at Urumqi river, mean annual temperature increased from –5.4 to 4.0 °C between 3408 and 2200 m asl.

Soil sampling and treatments. In all three localities, 6 mineral soil samples (40–60 mm below ground) were collected along the altitude gradient from random locations at each site between July 28 and August 15, 2001. Samples were kept in sterilized aluminum tins in the dark and cooled on ice until delivery to the laboratory and then refrigerated until processing. At the sampling site, dominant plants and communities characteristics were recorded.

Physical and chemical soil characterization. Soil pH was measured with a portable acidity meter in a soil–water (1 : 1, *M*/*V*) mixture. Air-dried soils were passed through a 100-mesh screen and soil organic carbon and total nitrogen determined using an element analyzer (*Perkin Elmer* 2400 II CHNS/O analyzer). Three replicates were performed for each sample.

Total number of soil microorganisms. DAPI staining was used to determine the total number of soil microbial cells. The total counts of bacteria and archaea were determined by the fluorescence *in situ* hybridization (FISH) technique. The probes ARCH915 (FAM-labeled) and EUB338 (Cy3-targeted) (*Sangon*, China) were synthesized. The 16S rRNA-labeled oligonucleotide sequence of probe EUB338 and ARCH915 was 5′-CY3–TGA GGA TGC CCT CCG TCG-3′ and 5′-FAM–GTG CTC CCC CGC CAA TTC CT-3′, respectively. The experimental protocol was described by Christensen *et al.* (1999). Three replicates were done for each sample.

Data analysis. One-way ANOVA was selected for testing significance levels among the same parameters at each location. Differences between the means were tested for their significance by using linear regression and variance analysis at $p < 0.05$.

RESULTS AND DISCUSSION

Total microorganisms. Generally, the counts of DAPI-stained cells were constant at different sites and varied from 1.8×10^9 to 8.6×10^9 cells per g soil dry mass (Table II, Fig. 2). The numbers of soil microbial cells were at Kalasi lake between 1.8×10^9 and 5.9×10^9. These counts were the lowest among the localities under study, the highest being the total numbers in the Urumqi river basin (Table II). On moving north, the counts of DAPI-stained cells gradually decreased from 8.0×10^9 and 6.4×10^9 to 2.9×10^9 (Table III) and the content of total organic carbon and total nitrogen was also lower. Kalasi lake soil was slightly acidic while Sangong river and Urumqi basin soils were nearly neutral to slightly alkaline. Additionally, Kalasi Lake has a lower temperature than the other localities (Table III). These results were in accordance with other studies and the differences among three locations could be attributed to differences in the soil organic carbon, total nitrogen and soil acidity. Staddon and Trevors (1998) demonstrated a decrease in microbial functional diversity with increasing latitude resulting in increasing environmental harshness, decreasing soil nutrient content, and increasing acidity. Meentemeyer and Berg (1986) observed poorer litter quality and lower atmospheric temperatures in high-latitude pine forest compared with low-latitude one and suggested that these factors would give rise to a lower diversity of the microbial communities across the latitude gradient. Microbial biodiversity in soil, ice, sediment, and water is positively correlated with the density of bacterial populations (Kaneko and Atlas 1977). Since soil microbial communities are often limited by carbon (Zak *et al.* 1994) or nitrogen (Zak *et al.* 1990), the lower population in Kalasi lake soil might indicate a lower biodiversity relative to localities situated down south. Therefore, the decrease in population may reflect increasing harshness of soil environment such as lower nutrient availability, and soil pH and low temperature.

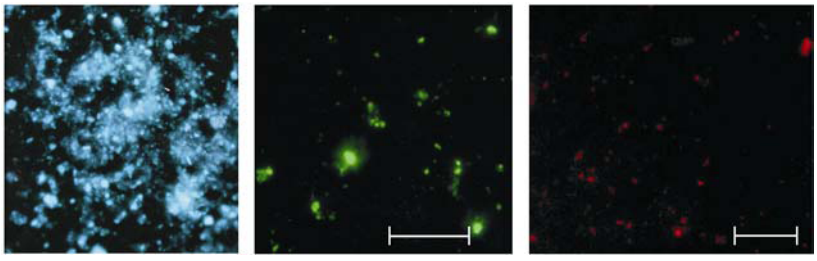

Fig. 2. The DAPI-stained cell counts (*left*), the number of archaea (*middle*), and bacteria (*right*) by 16S rRNA targeted fluorescence *in situ* hybridization (FISH) technique with the probe EUB338 and ARCH915, respectively; *bars* represent 10 μm.

Table II. The total DAPI-stained cell number and relative percentage of cells hybridized with specific probes EUB338 and ARCH915 for bacteria and archaea, and soil properties in different locations along an altitudinal gradient (means ± SD)*

Location	Elevation m	pH	Total cell number**	Bacteria† %	Archaea‡ %	Total N mg/g	Organic C %
Sangong river basin	1000	8.3	4.7 ± 2.2c	73.3 ± 3.2c	11.5 ± 1.0c	2.2 ± 0.2d	1.6 ± 0.3d
	1400	7.8	5.0 ± 1.6c	76.7 ± 2.3c	14.7 ± 1.6b	4.1 ± 0.9c	2.9 ± 0.5c
	1900	7.4	6.5 ± 1.1b	79.3 ± 4.8b	13.8 ± 1.4b	6.8 ± 1.0b	6.1 ± 0.6a
	2340	6.4	6.4 ± 1.5b	82.4 ± 2.9b	17.1 ± 2.1a	5.3 ± 0.5b	5.0 ± 0.4b
	3150	5.8	5.0 ± 1.6c	76.5 ± 1.4c	14.4 ± 1.5b	2.4 ± 0.3d	1.7 ± 0.7d
	3400	6.5	7.1 ± 1.9a	85.7 ± 1.7a	15.9 ± 2.2a	7.0 ± 0.8a	5.7 ± 0.6a
Urumqi river basin	1600	8.2	3.3 ± 1.4c	71.4 ± 2.2c	14.3 ± 1.0c	2.7 ± 0.5c	2.4 ± 0.2c
	2100	8.3	5.2 ± 0.8b	76.8 ± 1.8b	14.1 ± 1.2c	4.2 ± 0.5b	2.8 ± 0.4c
	2450	7.4	8.6 ± 1.0a	80.4 ± 1.7a	17.1 ± 3.0a	10.7 ± 1.2a	7.1 ± 1.1a
	2900	8.5	5.9 ± 1.3b	75.9 ± 1.1b	15.5 ± 0.9b	2.4 ± 0.6c	1.2 ± 0.1c
	3400	6.9	8.1 ± 0.9a	80.2 ± 2.1a	16.9 ± 1.2a	9.3 ± 1.9a	6.5 ± 1.4a
	3800	5.8	8.4 ± 0.7a	81.6 ± 1.8a	18.1 ± 0.5a	8.8 ± 0.8a	6.8 ± 0.9a
Kalasi lake	500	7.2	1.8 ± 0.5e	82.3 ± 1.1d	13.4 ± 1.2d	1.7 ± 0.1d	1.4 ± 0.2e
	950	6.4	2.8 ± 0.3d	82.4 ± 1.3c	15.4 ± 1.1c	1.9 ± 0.4d	1.8 ± 0.2e
	1500	6.4	5.9 ± 0.2a	73.5 ± 0.9a	20.6 ± 1.3a	8.7 ± 1.0a	7.4 ± 0.8a
	1700	5.4	3.6 ± 0.3c	73.9 ± 1.2a	16.6 ± 0.9a	3.2 ± 1.4c	3.2 ± 0.6d
	2100	5.3	3.6 ± 0.2c	78.7 ± 1.4c	17.9 ± 0.8c	2.1 ± 0.6d	1.7 ± 0.3e
	2350	5.6	4.7 ± 0.4b	72.2 ± 0.5b	18.8 ± 0.4b	5.9 ± 0.8b	4.5 ± 0.9c
	2500	6.0	4.8 ± 0.4b	76.6 ± 0.7b	20.0 ± 1.3b	6.2 ± 1.1b	5.9 ± 0.4b
	2680	5.8	4.0 ± 0.5b	69.4 ± 2.1c	18.8 ± 0.5c	5.6 ± 1.2b	5.1 ± 0.4b

*The different letters after numerals in each locality show a significant difference at $p < 0.05$ level with one-way ANOVA. The data on cell numbers were transformed with common logarithm, and the percentage of bacteria and archaea was transformed similarly after multiplying the total cell number.

**$\times 10^9$ per g soil dry mass; regression equation: $9.47 + 0.43\ C_{org}$, $p < 0.01$; $9.37 + 0.49\ N_{tot}$, $p < 0.01$.

†Regression equation: $2.86 + 0.003\ C_{org}$, $p < 0.05$; $2.81 + 0.004\ N_{tot}$, $p < 0.05$.

‡Regression equation: $2.14 + 0.10\ C_{org}$, $p < 0.05$; $2.13 + 0.11\ N_{tot}$, $p < 0.05$.

The number of bacteria and archaea in different vegetation types. FISH is regarded as a powerful tool in microbial ecology, permitting the detection of specifically stained single cells in different environments, determination of their phylogenetic relation and quantification of organisms in mixed communities without the need of isolation in pure cultures (cf. Amann *et al.* 1995). In this paper, probes EUB338 and ARCH915 were selected for detecting the number of soil bacteria and archaea, respectively (Tables II and III). Most of the soil microbial communities were bacterial, accounting for >69 % of the DAPI-stained cells whereas the total cell number of archaea was <20 %. The results were in accordance with those of Zak *et al.* (1994) who showed that bacteria formed the majority of soil microbial populations. Llobet-Brossa *et al.* (1998) reported that a large fraction (up to 73 %) of DAPI-stained cells in the uppermost layer of marine sediments was bacteria. The proportion of bacteria closely correlated with the input of organic material and the establishment of microbial communities.

Table III. The total DAPI-stained cell number, soil total nitrogen, organic carbon, and pH of Kalasi lake, Urumqi river, and Sangong river soils in Xinjiang (China)

Location	Total cell number[a]	Nitrogen, mg/g	Carbon, %	pH
Kalasi lake	2.91	4.17	3.27	6.5
Sangong river	6.39	4.59	3.66	7.0
Urumqi river	7.99	7.71	5.91	7.4

[a]$\times 10^9$ per g soil dry mass.

In each locality, the populations of bacteria and archaea collected at different elevations were statistically significantly different from each other and this difference correlated with the plant communities at each altitude (Table II). In general, the cells stained by FISH-specific dyes were more abundant in coniferous forest than in other vegetation types, sites with the desert vegetation having the least number of detec-

table microorganisms. Linear regression analysis indicated that the numbers of DAPI-stained cells, *i.e*. FISH-specific labeled microorganisms, were significantly and positively correlated with soil organic carbon and total nitrogen (Table II). The extant plant community is usually the main source for the input of C_{org} and N_{tot}, and the composition and diversity of the soil microbial community is obviously closely associated with the plant community. Due to the lack of plant cover and low rates of primary production, deserts are characterized by low C_{org} and N_{tot} levels, high soil pH and salinity; there are the causes of the lower population densities of bacteria and archaea. Lower temperature and soil pH in high-altitude alpine tundra and alpine meadow reletive to the medium altitude grassland and forest were apparently the main factors limiting the growth of microorganisms. Plant roots can release organic compounds, mainly polysaccharides, soluble secretions and cell lysates; coniferous forest, having a higher plant diversity and primary productivity than other vegetation types could therefore provide more organic material for soil microbes and support a more abundant population of soil microorganisms (Table II).

Research in this field suggested that the composition and diversity of the plant community influence the soil microbial community (Insam and Haselwandter 1989). At bryophyte-dominated sites, higher soil nutrient concentrations and higher production of easily degradable substrates are likely to maintain microbial activities (Ohtonen and Väre 1998). Likewise, Latour *et al.* (1996) have shown that the diversity of soil-borne populations of fluorescent *Pseudomonas* spp. was influenced by plant diversity.

This work was funded by *Knowledge-Innovation Projects of Chinese Academy of Sciences* (KZCX1-10-03, KZCX1-10-07), *The Key Laboratory of Ice Core and Cold Regions Environments, Chinese Academy of Sciences* (210 506), and the *Natural Science Foundation of China* (901 02003),

REFERENCES

AMANN R.I., LUDWIG W., SCHLEIFER K.H.: Phylogenetic identification and *in situ* detection of individual microbial cells without cultivation. *Microbiol.Rev.* **59**, 143–169 (1995).

BOGOEV V.M., KENAROVA A.E., VASILEV V.L., GYOSHEVA M.M.: Quantitative distribution of microbial biomass in the soil profile of a high-mountain grassy ecosystem. *Folia Microbiol.* **47**, 56–60 (2002).

BROUGHTON L.C., GROSS K.L.: Patterns of diversity in plant and soil microbial communities along a productivity gradient in Michigan old-field. *Oecologia* **125**, 420–427 (2000).

CHAPIN F.S. III, KORNER C.H.: Patterns, causes, changes and consequences of biodiversity in arctic and alpine ecosystems, in *Arctic and Alpine Biodiversity: Patterns, Causes, and Ecosystem Consequences, Ecological Studies, Vol. 113* (F.S. Chapin III, C.H. Korner, Eds). Springer-Verlag, Berlin 1995.

CHRISTENSEN H., HANSEN M., SØRENSEN J.: Counting and size classification of active soil bacteria by fluorescence *in situ* hybridization with an rRNA oligonucleotide probe. *Appl.Environ.Microbiol.* **65**, 1753–1761 (1999).

COUTEAUX M.M., BOTTNER P., BERG B.: Litter decomposition, climate and litter quality. *Tree* **10**, 63–66 (1995).

DERRY A.M., STADDON W.J., KEVAN P.G., TREVORS J.T.: Functional diversity and community structure of microorganisms in three arctic soils as determined by SCSU. *Biodivers.Conserv.* **8**, 205–221 (1999).

ELHOTTOVÁ D., SZILI-KOVÁCS T., TŘÍSKA J.: Soil microbial community of abandoned sand fields. *Folia Microbiol.* **47**, 435–440 (2002).

HEAD I.M., SAUNDERS J.R., PICKUP R.W.: Microbial evolution, diversity, and ecology: a decade of ribosomal RNA analysis of uncultivated microorganisms. *Microb.Ecol.* **35**, 1–21 (1998).

INSAM H., DOMSCH K.H.: Relationship between soil organic carbon and microbial biomass in chronosequences of reclamation sites. *Microb.Ecol.* **15**, 177–188 (1988).

INSAM H., HASELWANDTER K.: Metabolic quotient of the soil microflora in relation to plant succession. *Oecologia* **79**, 174–178 (1989).

KANDELER E., MARSCHNER P., TSCHERKO D., GAHOONIA T.S., NIELSEN N.E.: Microbial community composition and functional diversity in the rhizosphere of maize. *Plant & Soil* **238**, 301–312 (2002).

KANEKO T., ATLAS R.: Diversity of bacterial populations in the Beaufort Sea. *Nature* **270**, 596–599 (1977).

KAO C.M., CHEN S.C., CHEN Y.S., LIN H.M., CHEN Y.L.: Detection of *Burkholderia pseudomallei* in rice fields with PCR-based technique. *Folia Microbiol.* **48**, 521–524 (2003).

LATOUR X., PHILIPPOT L., CORBERAND T., LEMANCEAU P.: The establishment of an introduced community of fluorescent pseudomonads in the soil and in the rhizosphere is affected by the soil type. *FEMS Microbiol.Ecol.* **30**, 163–170 (1996).

LLOBET-BROSSA E., ROSSELLÓ-MORA R., AMANN R.: Microbial community composition of Wadden Sea sediments as revealed by fluorescence *in situ* hybridization. *Appl.Environ.Microbiol.* **64**, 2691–2696 (1998).

MARILLEY L., ARAGNO M.: Phylogenetic diversity of bacterial communities differing in degree of proximity of *Lolium perenne* and *Trifolium repens* roots. *Appl.Soil Ecol.* **13**, 127–136 (1999).

MEENTEMEYER V., BERG B.: Regional variation in rate of mass loss of *Pinus sylvestris* needle litter in Swedish pine forests as influenced by climate and litter quality. *Scand.J.Forest Res.* **1**, 167–180 (1986).

MUYZER G., SMALLA K.: Application of denaturing gradient gel electrophoresis (DGGE) and temperature gradient gel electrophoresis (TGGE) in microbial ecology. *Antonie van Leeuwenhoek* **73**, 127–141 (1998).

OHTONEN R., VÄRE H.: Vegetation composition determines microbial activities in a boreal forest soil. *Microb.Ecol.* **36**, 328–335 (1998).

STADDON W.J., TREVORS J.T., DUCHESNE L.C., COLOMBO C.A.: Soil microbial diversity and community structure across a climatic gradient in western Canada. *Biodivers.Conserv.* **7**, 1081–1092 (1998).

STEPHAN A., MEYER A.H., SCHMID B.: Plant diversity affects culturable soil bacteria in experimental grassland communities. *J.Ecol.* **88**, 988–998 (2000).

SWIFT M.J., ANDRÉN O., BRUSSAARD L., BRIONES M., COUTEAUS M.M., EKSCHMITT K., KJOLLER A., LOISEAU P., SMITH P.: Global change, soil biodiversity, and nitrogen cycling in terrestrial ecosystems: three case studies. *Global Change Biol.* **4**, 729–744 (1998).

TIEDJE J.M., ASUMING-BREMPONG S., NÜSSLEIN K., MARSH T.L., FLYNN S.J.: Opening the black box of soil microbial diversity. *Appl. Soil Ecol*. **13**, 109–122 (1999).

VESELOVA M., KHOLMECKAYA M., KLEIN S., VORONINA E., LIPASOVA V., METLITSKAYA A., MAYATSKAYA A., LOBANOK E., KHMEL I., CHERNIN L.: Production of *N*-acylhomoserine lactone signal molecules by Gram-negative soil-borne and plant-associated bacteria. *Folia Microbiol.* **48**, 794–798 (2003).

WAHLSTRÖM G., DANILOV R.A.: Phytoplankton successions under ice cover in four lakes located in North-Eastern Sweden: effects of liming. *Folia Microbiol.* **48**, 379–384 (2003).

WARDLE D.A., VERHOEF H.A., CLARHOLM M.: Trophic relationships in the soil microfood-web: predicting the responses to a changing global environment. *Global Change Biol.* **4**, 713–727 (1998).

WHITTAKER R.H.: *Communities and Ecosystems*. MacMillan Publisher, New York 1975.

ZAK D.R., GRIGAL D.R., GLEESON S., TILMAN D.: Carbon and nitrogen cycling during old-field succession: constraints on plant and microbial biomass. *Biogeochemistry* **11**, 111–129 (1990).

ZAK D.R., TILMAN D., PARMENTER R.R., RICE C.W., FISHER F.M., VOSE J., MILCHUNAS D., MARTIN C.W.: Plant production and soil microorganisms in late-successional ecosystems: a continental scale study. *Ecology* **75**, 2333–2347 (1994).

ZHANG X.J., YAO T.D., MA X.J., WANG N.L.: Microorganisms in a high altitude glacier ice in Tibet. *Folia Microbiol.* **47**, 241–246 (2002).