ANALYSIS OF BACTERIAL DIVERSITY IN ARCTIC SOIL AND SEDIMENT SAMPLES FROM NY-ÅLESUND, NORWAY USING DENATURING GRADIENT GEL ELECTROPHORESIS

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ABSTRACT

Bacterial diversity in soil and sediment samples from eighteen sites in and around Dasan Station, Ny-Ålesund, Norway was analyzed using Denaturing Gradient Gel Electrophoresis (DGGE), and was correlated with the environmental variables. Samples analysed included terrestrial soils, beach soil, periglacier soils, inland lake bank sediments, melt lake sediments, and marine sediments.

Spearman rank order correlation showed significant correlations (*P* = 0.001) between conductivity (*r* = 0.470) and pH (*r* = 0.294) with the bacterial diversity. Soil from the tundra site (sample 17) appeared to have the most diverse bacterial community while inland lake bank sediment from the freshwater site (sample 35) showed the least diverse bacterial community. Non-metric multidimensional scaling plots (nMDS) and Hierarchical cluster analysis showed that the bacterial community structure was eventually clustered into two groups: non-marine and marine samples.

Out of 28 DGGE bands that were sequenced, nine sequences were related to unclassified bacteria, four sequences each were related to *Bacteroidetes* and *β-proteobacteria,* two each were related to *Cyanobacteria*, *Firmicutes* and *Fusobacteria*, and one each was related to *Acidobacteria*, *ε-proteobacteria*, *δ-proteobacteria*, *Fibrobacteres* and *Nitrospira*. Despite unclassified bacteria, *Bacteroidetes* were dominant in marine (22.45%) and beach samples (28.55 %) while *β-proteobacteria* were the dominant group in terrestrial (15.85 %) and melt lake (21.05 %) samples. On the other hand, the inland lake bank samples were dominated (14.81 %) by *Cyanobacteria*, *Fibrobacteres*, *Firmicutes* and *β-proteobacteria*. Where as, the periglacier samples were dominated (21.05 %) by *Bacteroidetes* and *β-proteobacteria*.

Phylogenetic analysis of sequences of well-defined DGGE bands displayed four distinct clades, which consisted of (i) *Bacteroidetes* and *Cyanobacteria*; (ii) *Firmicutes*, *Nitrospira*, *Fibrobacteres*, *Acidobacteria*, *ε-proteobacteria* and *δ-proteobacteria*; (iii) *β-proteobacteria*; and (iv) *Fusobacteria*. There was a distinct clade of uncultured representatives that were not closely related to any known GenBank sequences. This probably represent a potential gene pool of novel species which yet to be cultured or identified.

ABSTRAK

Kepelbagaian bakteria yang berada di dalam tanah dari lapan belas tempat di sekitar Stesen Dasan, Ny-Ålesund, Norway telah dianalisis dengan teknik Denaturing Gradient Gel Electrophoresis (DGGE), dan dihubungkaitkan dengan data pembolehubah persekitarannya. Sampel yang dikaji termasuk tanah daratan, pasir pantai, tanah sungai ais, endapan di tepi tasik, endapan tasik ais cair, dan endapan laut.

Korelasi Spearman rank order telah menunjukkan nilai hubungan bermakna (P = 0.001) di antara kepelbagaian bakteria dengan konduktiviti (r = 0.470) dan pH sampel (r = 0.294). Non-metric multidimensional scaling plots (nMDS) dan analisis Hierarchical cluster telah menunjukkan komuniti bacteria terbahagi kepada dua kumpulan: sample bukan laut dan laut. Tanah dari padang kutub (sampel 17) telah memaparkan nilai kepelbagaian bakteria yang tertinggi manakala tanah dari endapan tasik air tawar (sampel 35) telah memaparkan nilai kepelbagaian bakteria yang paling rendah.

Daripada jumlah 28 jujukan gen yang diterima, sembilan jujukan gen berkaitan dengan unclassified bacteria, empat dengan *Bacteroidetes* dan *β-proteobacteria*, dua yang setiapnya dengan *Cyanobacteria*, *Firmicutes*, dan *Fusobacteria*; serta satu yang setiapnya dengan *Acidobacteria*, *ε-proteobacteria*, *δ-proteobacteria*, *Fibrobacteres*, dan *Nitrospira*. *Bacteroidetes* merupakan populasi utama di dalam sampel endapan laut (22.45%) dan pasir pantai (28.55 %) manakala *β-proteobacteria* merupakan populasi utama di dalam sampel tanah daratan (15.85 %) dan endapan tasik ais cair (21.05 %). Di samping itu, endapan di tepi tasik didominasi (14.81 %) oleh *Cyanobacteria*, *Fibrobacteres*, *Firmicutes* dan *β-proteobacteria*. Manakala tanah sungai ais pula didominasi (21.05 %) oleh *Bacteroidetes* dan *β-proteobacteria*.

Analisis filogenetik telah memaparkan jujukan gen dalam jalur DGGE tergolong dalam empat kumpulan nyata yang terdiri daripada (i) *Bacteroidetes* dan *Cyanobacteria*; (ii) *Firmicutes*, *Nitrospira*, *Fibrobacteres*, *Acidobacteria*, *ε-proteobacteria* dan *δ-proteobacteria*; (iii) *β-proteobacteria*; serta (iv) *Fusobacteria*. Terdapat satu kumpulan jalur DDGE yang tidak berkait rapat dengan apa-apa bakteria spesies di dalam GenBank BLAST. Ini mungkin mewakili satu kumpulan bakteria baru yang belum dikenali dan perlu dikaji.

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ABBREVIATIONS

% : percent
= : equal to
> : more than
< : less than
℃ : degree Celsius
µg : microgram
µl : microlitre
µS : microsiemens
A_{260}/A_{230} : ratio of UV absorbance at 260nm and 230nm
A_{260}/A_{280} : ratio of UV absorbance at 260nm and 280nm
AGE : agarose gel electrophoresis
ANOSIM : analysis of similarity
BEST : the biota and/or environment matching
Bis : N,N'-methylenbisacrylamide
BLAST : basic local aligned search tool
bp : base pairs
BPB : bromophenol blue
DGGE : denaturing gradient gel electrophoresis
DNA : deoxyribonucleic acid
dNTP : deoxyribonucleoside triphosphate
E : East
EC : electric conductivity
EDTA : ethylenediaminetetraacetate acid
EtBr : ethidium bromide
F : forward
g : gram
H′ : Shannon’s diversity index
Inc : Incorporate

IRS	: inhibitor removal solution
L	: litre
M	: molar
MEGA	: molecular evolutionar genetics analysis
mg	: milligram
min	: minute
ml	: millilitre
mM	: millimolar
N	: North
NaCl	: sodium chloride
NJ	: neighbor-joining
nm	: nanometer
nMDS	: non-metric multidimensional scaling
PCR	: polymerase chain reaction
R	: reverse
r	: global value of Spearman rank order correlation
rcf	: relative centrifugal force
rDNA	: ribosomal deoxyribonucleic acid
RNA	: ribonucleic acid
rRNA	: ribosomal ribonucleic acid
s	: second
sdH_2O	: sterile distilled water
SSU	: small subunit
TAE	: tris acetate ethylenediaminetetraacetate acid
Taq	: *Thermus aquaticus*
UV	: ultraviolet
V	: volt
v/v	: volume per volume
w/v	: weight per volume

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