

Bacterial diversity of Drass, cold desert in Western Himalaya, and its comparison with Antarctic and Arctic

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Abstract Drass is the coldest inhabited place in India and the second coldest, inhabited place in the world, after Siberia. Using the 16S rDNA amplicon pyrosequencing, bacterial diversity patterns were cataloged across the Drass cold desert. In order to identify the ecotype abundance across cold desert environment, bacterial diversity patterns of Drass were further compared with the bacterial diversity of two other cold deserts, i.e., Antarctic and Arctic. *Acidobacteria*, *Proteobacteria*, *Actinobacteria*, *Bacteroidetes*, *Cyanobacteria* and *Gemmatimonadetes* were among the highly abundant taxonomic groups present across all the three cold deserts and were designated as the core phyla. However, *Firmicutes*, *Nitrospirae*, *Armatimonadetes* (former candidate division OP10), *Planctomycetes*, *TM7*, *Chloroflexi*, *Deinococcus-Thermus*, *Tenericutes* and candidate phyla *WS3* were identified as rare phyla in Drass, Antarctic and Arctic samples. Differential abundance patterns were also computed across all the three samples, i.e., *Acidobacteria* (32.1 %) were dominant in Drass and *Firmicutes* (52.9 ± 17.6 %) and *Proteobacteria* (42 ± 1.3 %) were dominant in Antarctic and Arctic reference samples, respectively. Alpha diversity values Shannon's (*H*) and Simpson's (1-D) diversity indices were highest for Antarctic samples, whereas richness estimators (ACE and Chao1) were maximum for Drass soil suggesting greater species

richness in bacterial communities in Drass than the Antarctic and Arctic samples.

Keywords Bacterial diversity · Cold desert · Pyrosequencing · Drass · Himalayas

Introduction

The Himalayan cold deserts are characterized by a fragile ecosystem and a complex climate due to dramatic seasonal shifts in physical and biochemical properties. Bordered by the Karakoram chain of mountains in the north and the Himalayas in the south (Rawat and Adhikari 2005), Ladakh represents a high-altitude cold desert. The Karakoram range blocks most of the monsoon bearing clouds, making the region arid (Namgail 2009). Based on the cloning-based cultivation-independent methods (Shivaji et al. 2011; Srinivas et al. 2011; Kistler et al. 2013), recent studies have reported the bacterial diversity of Himalayan regions. Though cloning-based cultivation-independent approaches overcome the limitations of cultivation-based methods, owing to the methodology-based bias, e.g., PCR and sequencing depth (Vaz-Moreira et al. 2011), these techniques are not efficient enough to catalog the rare microbial taxons. High-throughput sequencing of total environmental DNA or marker genes, e.g., 16S rRNA, circumvents cloning by taking advantage of a highly efficient in vitro DNA amplification method. This approach provides deeper insights into the taxonomical and functional dynamics of the in situ microbial diversity (Dowd et al. 2008; Rhoads et al. 2012; Kaur et al. 2015).

The primary goal of the present study was to explore the bacterial community dynamics of cold desert of Drass and its further comparison with other cold deserts, e.g.,

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Antarctic and Arctic. Comparative analysis has been done to identify the differentially abundant bacterial taxa in cold deserts with different geographical conditions, i.e., ecotypes. Here, we provide a basic framework to analyze the bacterial diversity patterns enriched across cold desert environment. However, this framework would be benefitted via deeply sequenced longitudinal time series datasets.

Materials and methods

Sampling site

Drass is a small town in the Kargil district of Ladakh region (J&K, India) at 34.45°N, 75.77°E with an average elevation of 3280 meters (10,764 feet). Soil samples ($n = 10$) were collected from different locations in the Drass region. One centimeter of the surface soil was removed with a sterile spatula before collection of soil samples. Soil was collected with sterile spatula and transferred into sterile polythene bags. The temperature of the soil ranged from 10 to 15 °C at the time of sampling. The soil samples were transported to the laboratory under ice and then stored at -20 °C (Shivaji et al. 2011).

Physiochemical soil analysis

Soil samples were thawed, air-dried and passed through a 2-mm sieve and pooled as one composite sample and sent for analysis to QC research lab IIIM Jammu, India, for analysis of pH, water holding capacity and various metal ions concentration in the soil. All determinations were conducted using triplicate samples.

Metagenomic DNA extraction and amplification

Manual metagenomic DNA extraction protocols developed by Zhou et al. 1996; Wechter et al. 2003; Brady 2007; Amorim et al. 2008; Pang et al. 2008; Liles et al. 2009; Inceoglu et al. 2010 were applied for environmental DNA extraction. DNA extracted from multiple methods ($n = 3$) was pooled and diluted (1/100 dilutions) with final concentration of 20 ng/ μ l and used as template for PCR amplification. Small region (V1–V3) of the 16S rRNA gene was amplified from the total soil DNA by PCR using universal primers set 28F and 519 R (Nossa et al. 2010; Fan et al. 2012).

Pyrosequencing

Bacterial tag-encoded FLX amplicon pyrosequencing (bTEFAP) (Dowd et al. 2008) was performed at the Research and Testing Laboratory, Lubbock, TX, USA (www.researchandtesting.com).

Quality filtering and phylogenetic analysis

Raw sequence data were processed and analyzed using the Quantitative Insights into Microbial Ecology (QIIME version 1.5.0) pipeline, with default settings (Caporaso et al. 2010). Reads were removed after further analysis if at least one of the following criteria was met: (1) the presence of homopolymers with more than 8 bp and (2) length shorter than 200 bp. Quality-filtered reads were denoised to remove sequencing errors by flowgram clustering. The resulting (trimmed and clean) sequences were then filtered of any non-bacterial ribosome sequences set at default parameters (Gontcharova et al. 2010). The 454 sequence data comprise both identical reads and reads that start at the same position in the genome but have different lengths or vary slightly due to pyrosequencing errors. These erroneous reads can lead to an overestimation of several operational taxonomic units in a sample. Uchime tool was used to remove the chimeric sequences (Edgar et al. 2011). Quality trimmed sequences were clustered into operational taxonomic units (OTUs) using UClust (Edgar 2010) with a cutoff value of 99 % sequence identity. Candidate OTUs were assigned to phylogeny using RDP (Cole et al. 2009; Krobe et al. 2009) scheme set at 80 % confidence value. Rarefaction curves were calculated by using RDP pyrosequencing pipeline (Cole et al. 2009).

Reference datasets

A total of four reference datasets were obtained from NCBI. Two reference datasets were selected randomly from the Antarctic study with accession nos. SRX206452 and SRX206985, and remaining two reference datasets were selected randomly from the Arctic study with accession no SRX017110. The source of Antarctic and Arctic samples was from Grove Mountains, East Antarctic and Foreland of MidreLoven glacier.

Statistical analysis

For each metagenome, a subset of 1000 candidate OTUs were randomly selected to avoid the biases caused by differential sampling size (Kirchman et al. 2010). Inter-sample alpha diversity comparisons (Kirchman et al. 2010) were performed using a Vegan package (Oksanen et al. 2013). Sequences were aligned for phylogenetic reconstruction using MAFFT algorithm that appeared in the version 1.7.2 of UGENE as the External Tool. Phylogenetic trees were built from the aligned sequence profiles using FASTTREE 2.1 (Price et al. 2010). Distance matrices were constructed from each phylogeny, and pairwise Mantel test (1000 permutations, two tailed: p value) was performed using PASSAGE-2 (Rosenberg and Anderson 2011). To assess the

uncertainty in hierarchical cluster analysis over samples, bootstrap resampling (10,000 iterations) analysis implemented in Pvcust (Felsenstein 1985) was applied. Pvcust provides the bootstrap probability (BP) value from the ordinary bootstrap resampling (Felsenstein 1985) and the approximately unbiased (AU) probability value from multi-scale bootstrap resampling (Shimodaira and Hasegawa 2001; Suzuki and Shimodaira 2006). Cluster dendrogram was constructed with AU/BP values (%). A heat map showing the pairwise Pearson coefficient correlations among the samples was plotted using heat map function implemented in R.

Nucleotide sequence accession number

The 16S rRNA gene sequences derived from pyrosequencing of Drass soil have been deposited in the European Nucleotide Archive under accession number PRJEB5191.

Results

Soil of Drass cold desert is sandy, slightly alkaline (pH 7.4), nutrient poor with 0.72 % organic content and 12 ds/m electric conductivity (EC). Physicochemical properties of composite soil sample from Drass are given in Table 1.

Only three DNA extraction protocols, developed by Zhou et al. (1996); Wechter et al. (2003) and Pang et al. (2008), worked efficiently on the soil of Drass. DNA extracted using these three protocols were pooled and used as a template for PCR amplification of V1–V3 region of 16S rDNA. The amplicons were pyrosequenced, and 3000 sequence reads were generated. Downstream quality filtering resulted in 2819 high-quality sequences with average read length of ≥ 200 bp. Phylogenetic analysis revealed *Acidobacteria*, *Proteobacteria* and *Actinobacteria* as highly abundant in the soil of Drass representing 39 % of the total bacterial sequences. In addition, *Chloroflexi*, *Bacteroidetes*, *Verrucomicrobia*, *Planctomycetes*, *Firmicutes*,

Nitrospira, *Armatimonadetes* (former candidate division (OP10), *Gemmatimonadetes* and *Cyanobacteria* were also identified at the relatively low abundance (<10 %), along with candidate phyla *WS3* and *TM7* and several unclassified bacteria (Fig. 1).

Comparative account of bacterial diversity across Drass, Antarctic and Arctic soil samples

The 16S rRNA gene sequences from the present study (Drass soil) and those retrieved from NCBI (Arctic and Antarctic soil) were clustered at 99 % sequence identity (Egge et al. 2013) (Table 2). A significant proportion (2.2, 2.8 and 1.9 %) of the total sequences from Drass and Antarctic (ANT1 and ANT2) samples was observed as chimeric (Uchime), and samples were filtered before clustering. No chimeric sequences were detected in Arctic samples. The rarefaction curves constructed at the 1 % genetic distance exhibited a steeper slope for the Drass soil than Antarctic and Arctic soil samples, hence demonstrating a greater genetic richness in the Drass soil (Fig. 2a). Drass sample, however, needs extensive sampling to represent the taxonomic diversity. Furthermore, Shannon's (*H*) and Simpson's diversity indices were highest for Antarctic samples, whereas ACE and Chao1 were maximum for Drass soil suggesting higher species richness across Drass soil sample than the Antarctic and Arctic soil samples (Table 3). Beta-diversity and hierarchical clustering analysis revealed that the total bacterial community structure in the Drass is more similar to Arctic than Antarctic soil samples (Fig. 2b–d).

Abundant versus rare OTU analysis

Core/abundant phyla in Drass soil, Antarctic samples and Arctic samples

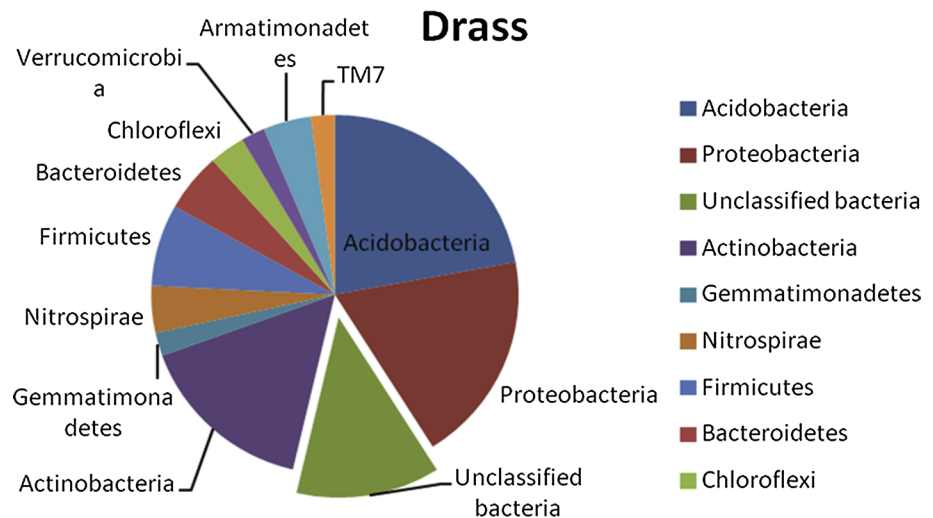
OTUs represented in all the five samples, i.e., Drass (DR), Antarctic (ANT1 and ANT2) and Arctic (ARC1 and

Table 1 Physicochemical analysis of soil samples

Characteristic	Drass (DR) ^a	Antarctic (ANT1 and ANT2)	Arctic (ARC1 and ARC2)
Texture	Sandy, coarse	–	–
Color	Greyish brown	–	–
pH	7.4 ± 0.15	6–6.7	6–6.7
Water content (%)	19 ± 0.3	–	–
EC (dS/m)	12 ± 0.5	–	–
Organic carbon (%)	0.72 ± 0.05	–	–
Nitrogen (%)	0.54 ± 0.05	–	–
Available K (kg/ha)	72 ± 0.82	–	–
Available P (kg/ha)	50 ± 1.1	–	–

^a Experiments were conducted in triplicates and the data was expressed as mean ± SD

Fig. 1 Pie chart showing phylogenetic diversity (phylum level) of 16S rRNA gene sequences obtained from bTEFAP analysis of Drass metagenome



ARC2), were collectively termed as core phyla (Serkebaeva et al. 2013) affiliated with the *Acidobacteria*, *Proteobacteria*, *Actinobacteria*, *Verrucomicrobia*, *Bacteroidetes*, *Cyanobacteria* and *Gemmatimonadetes* (Fig. 3a). The relative percentage abundance of phyla, class and orders mentioned in the below paragraph is given in Table 2.

Interestingly, *Acidobacteria* dominated the bacterial diversity of Drass sample with Gp6, Gp4 and GP7 being the abundant classes. Relative abundance patterns of Class Gp6 and Gp4 was lower in the Antarctic and Arctic samples with GP7 as entirely absent. Proteobacteria were abundant across all the three sites; however, they were comparatively more predominant in Arctic soil samples than Drass and Antarctic samples. Order *Rhizobiales*, *Sphingomonadales*, *Caulobacterales* of the *Alphaproteobacteria*, *Burkholderiales* of the *Betaproteobacteria* and *Pseudomonadales* and *Xanthomonadales* of the *Gammaproteobacteria* were represented among all the three sites. *Enterobacteriales* (*Gammaproteobacteria*) were represented among Antarctic samples only. Order *Myxococcales* (*Deltaproteobacteria*) were not represented in Antarctic and Arctic subsamples.

There were no significant differences in the abundance of *Actinobacteria* across Drass, Antarctic and Arctic soil samples. Unclassified order *Actinomycetales* was present across all the soil samples. However, order *Solirubrobacterales* and *Acidimicrobiales* were observed in Drass only and Arctic soil samples. Interestingly, *Gaiellales* were detected in Arctic samples only. At genus level, *Aciditerrimonas*, *Conexibacter*, *Patulibacter*, *Rubrobacter*, *Propionibacterium*, *Marmoricola*, *Nocardioides*, *Aeromicrobium*, *Arthrobacter*, *Leifsonia*, *Amycolatopsis*, *Corynebacterium* and *Solirubrobacter* were uniformly distributed in the Drass soil sample. *Bacteroidetes* were less abundant in Drass than Antarctic and Arctic samples. Order *Sphingobacteriales* was commonly represented across Drass, Antarctic and Arctic samples. However, *Cytophagales* were detected across Arctic samples only.

Members of the *Verrucomicrobia* were more abundant in Drass than the Antarctic soil and Arctic soil samples. Order *Opitutales* was present in higher abundance across Drass sample, but not detected in the Antarctic and Arctic samples. Similarly, Order *Verrucomicrobiales* not detected in Drass was relatively abundant in Antarctic samples and detected in Arctic samples. Phylum *Cyanobacteria* were significantly less abundant in Drass than in Antarctic and Arctic samples. Class *Chloroplast* was abundant across Antarctic samples, and class *Cyanobacteria* was abundant across Arctic samples. Phylum *Gemmatimonadetes* represented by single genus, i.e., *Gemmatimonas*, was predominant across Drass sample and relatively very less in Antarctic and Arctic samples.

Rare OTUs across Drass, Arctic and Antarctic samples

The OTUs detected in two samples only were referred to as rare OTUs (Serkebaeva et al. 2013). *Firmicutes*, *Nitrospirae*, *Armatimonadetes* (Former candidate division OP10), *Planctomycetes*, *TM7*, *Chloroflexi*, *Deinococcus-Thermus*, *Tenericutes* and Candidate *WS3* were distinguished as rare genera (Fig. 3b). *Brevibacillus* was the most abundant genera of *Firmicutes* present in Drass sample. Similarly, *Leuconostoc*, *Tumebacillus* and *Staphylococcus* were the most abundant genera (rare) present across Antarctic samples. The overall proportion of phyla *Nitrospira*, *Planctomycetes*, *Chloroflexi* and *Armatimonadetes* was significantly higher in Drass than in Antarctic and Arctic samples. *Tenericutes* were detected in Antarctic samples only and Candidate phylum *WS3* detected in Drass only in the present study.

Discussion

Extreme conditions are the characteristics of cold desert alpine soils due to temperature fluctuations, and this holds

Table 2 Percentage of core phyla and rare phyla obtained from metagenome of Drass, Antarctic (ANT1 and ANT2), Arctic (ARC1 and ARC2)

Samples	DR (%)	ANT1 (%)	ANT2 (%)	ARC1 (%)	ARC2 (%)
No. of phyla	14	12	8	13	12
Core phyla					
<i>Acidobacteria</i>	32.1	<1	<1	4.3	4.2
<i>Gp4</i> (class)	8.66	<1	–	1.3	1.87
<i>Gp6</i> (class)	14.8	–	–	<1	<1
<i>Gp7</i> (class)	3.09	–	–	–	–
<i>Proteobacteria</i>	21.4	14.8	31.8	40.7	43.3
<i>Deltaproteobacteria</i>	5	<1	<1	–	–
<i>Myxococcales</i> (order)	3.31	–	–	–	–
<i>Betaproteobacteria</i>	4.74	3.26	10.3	3.12	3.3
<i>Burkholderiales</i> (order)	3.08	3.06	7.8	2.5	2.6
<i>Alphaproteobacteria</i>	9.54	3.38	6.7	32	38.1
<i>Rhizobiales</i> (order)	4.11	1.45	2.3	7.5	10.35
<i>Sphingomonadales</i> (order)	1	1	2.3	13.5	12.72
<i>Caulobacteriales</i> (order)	<1	<1	1.1	2.08	2.2
<i>Gammaproteobacteria</i>	9.43	7.8	14.9	36/1535	27/1226
<i>Pseudomonadales</i> (order)	6.17	<1	<1	1	<1
<i>Enterobacteriales</i> (order)	–	4.8	10.5	–	–
<i>Xanthomonadales</i> (order)	1.27	2.42	3.6	<1	<1
<i>Actinobacteria</i>	6.6	2	8.5	11.2	12.9
<i>Actinomycetales</i> (order)	2.1	2	8.5	7.4	9.9
<i>Solirubrobacteriales</i> (order)	1.1	–	–	1.2	1
<i>Acidimicrobiales</i> (order)	<1	–	–	2.02	13
<i>Gaiellales</i> (order)	–	–	–	<1	<1
<i>Verrucomicrobia</i>	1.7	1	1	<1	<1
<i>Spartobacteria</i> (class)	<1	–	–	–	–
<i>Opitutae</i> (class)	1.3	–	–	–	–
<i>Verrucomicrobiae</i> (class)	–	1	1	<1	<1
<i>Bacteroidetes</i>	4.8	6	7.3	18.1	15.4
<i>Sphingobacteriales</i> (order)	2.6	3.5	4.1	9.3	8.6
<i>Flavobacteriales</i> (order)	<1	2.3	2.8	1	<1
<i>Cytophagales</i> (order)	–	–	–	4.6	3.3
<i>Cyanobacteria</i>	<1	3.8	10.7	6.6	5.05
<i>Cyanobacteria</i> (class)	–	–	–	6.6	5.05
<i>Chloroplast</i> (class)	<1	3.8	10.7	–	–
<i>Gemmatimonadetes</i>	4.1	<1	<1	<1	<1
<i>Gemmatimonas</i> (genus)	4.1	<1	<1	<1	<1
Rare phyla					
<i>Firmicutes</i>	2.5	70.5	35.3	–	–
<i>Nitrospira</i>	1.3	<1	<1	–	–
<i>Armatimonadetes</i>	1.1	–	–	<1	<1
<i>Planctomycetes</i>	<1	–	–	–	–
<i>TM7</i>	<1	–	–	<1	<1
<i>Deinococcus-Thermus</i>	–	<1	<1	<1	<1
<i>Chloroflexi</i>	1.5	<1	–	–	<1
<i>Tenericutes</i>	–	<1	1.9	–	–
<i>Candidate WS3</i>	<1	–	–	–	–

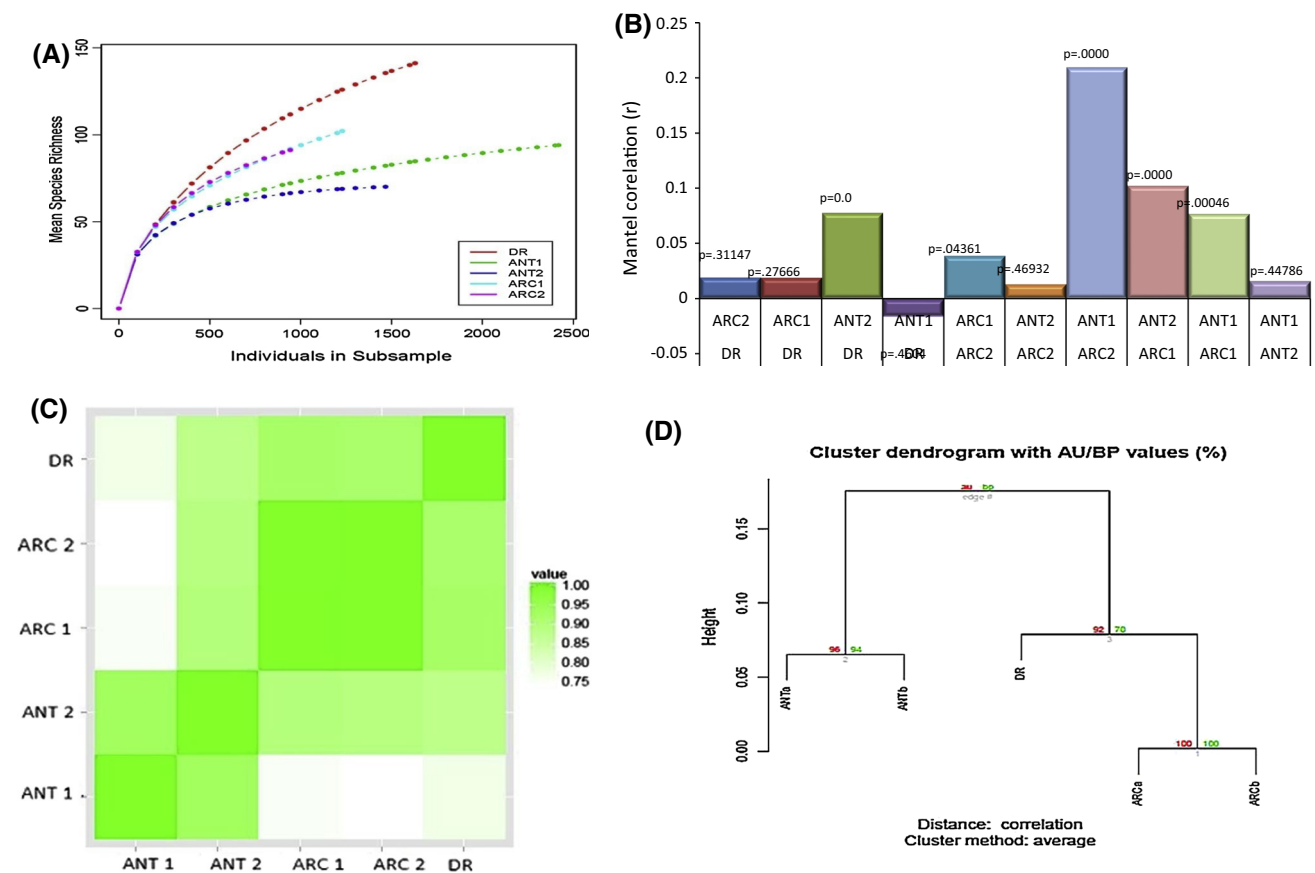


Fig. 2 **a** Rarefaction analysis performed on samples Drass (DR), Antarctic 1 (ANT1), Antarctic 1 (ANT2), Arctic 1 (ARC1) and Arctic 2 (ARC2). **b** Phylogenetic correlation of microbial communities of Drass, Antarctic and Arctic. A subset of 1000 randomly selected OTUs from each metagenome was used to construct an Euclidean distance matrix. Matrices were pairwise compared using Mantel test (1000 permutation, 0.05 as standard P value), and Pearson correlation values were calculated. Symbols used: Drass (DR), Antarctic 1 (ANT1), Antarctic 1 (ANT2), Arctic 1 (ARC1) and Arctic 2 (ARC2). **c** Heat map: Pearson correlation distances, ranging from 0 to 2, were normalized to a scale of 0–1, where 1 represents the furthest distance,

or the least similar samples. Symbols used: Drass (DR), Antarctic 1 (ANT1), Antarctic 1 (ANT2), Arctic 1 (ARC1) and Arctic 2 (ARC2). **d** To assess the uncertainty in hierarchical cluster analysis over samples Drass (DR), Antarctic 1 (ANT1), Antarctic 1 (ANT2), Arctic 1 (ARC1) and Arctic 2 (ARC2). Bootstrap resampling (10,000 iterations) was applied via the R package Pvcust. The uncentered Pearson correlation is used as the distance metric with average linkage. The numbers above each edge show the probability of nodes below that edge occurring as a cluster in resampled trees, via ordinary bootstrap resampling (BP, green) or multi-scale bootstrap resampling (AU, red) (color figure online)

true for Drass as well. To our knowledge, this is the first report on the bacterial diversity (richness and distribution) of Drass by high-throughput tag-encoded FLX amplicon pyrosequencing and its comparison with the bacterial diversity reported from other two cold deserts, Antarctic and Arctic based on retrieved sequence sets from the NCBI.

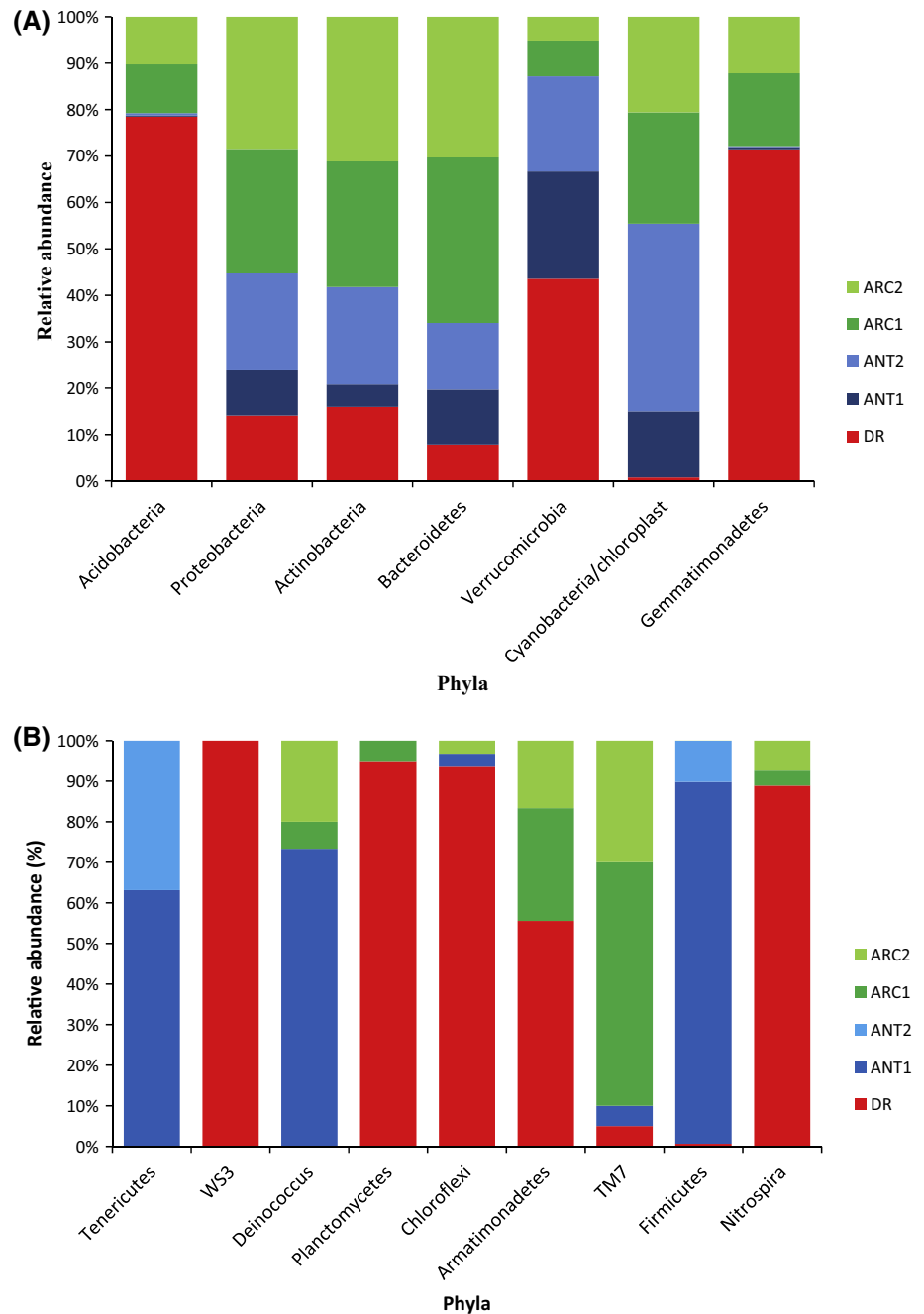
Low organic content in Drass is due to lack of vegetation, poor microbial activities, coarse sediments and low humus content (Kastovska et al. 2005; Rawat and Adhikari 2005; Sagwal 1997; Charan et al. 2013). Organic carbon content, soil texture and salinity have a strong influence on the microbial community diversity (Fierer et al. 2003, 2007). Multiple DNA extraction protocols were employed since no single method of metagenomic DNA isolation is efficient enough to represent all the bacterial diversity

(Delmont et al. 2011). Sequence analysis revealed the predominance of *Acidobacteria* in Drass soil despite slightly alkaline pH although expect to be more in Antarctic and Arctic soil with acidic pH (6–6.7) (Li et al. 2003; Kastovska et al. 2005). This could be due to low carbon availability, soil organic carbon (OC), soil C/N ratio, as the abundance of *Acidobacteria* and the organic carbon content of the soil have been negatively co-related (Fierer et al. 2007; Pointing et al. 2009; Zeglin et al. 2011; Lee et al. 2012; Naether et al. 2012; Bottos et al. 2014; Niederberger et al. 2015). In addition, the Class Gp6 and Gp4 of *Acidobacteria* are reported to be present at high pH (Jones et al. 2009) and that could be the reason for their abundance in Drass in comparison with the Antarctic and Arctic samples.

Table 3 Diversity indices among bacterial communities in Drass (DR), Antarctic 1 (ANT1), Antarctic 1 (ANT2), Arctic 1 (ARC1) and Arctic 2 (ARC2) estimated by vegan tool

Diversity indices	DR	ANT1	ANT2	ARC1	ARC2
OTU	1818	8276	1893	1536	1227
Shannon–Wiener index (H') nats	3.181599	3.247597	3.250028	3.110686	3.138125
Simpson (1-D)	0.8951769	0.9228484 0	0.93353020	0.8932482	0.8990074
S.chao1	194.000000	115.230769	71.666667	160.57143	116.375000
se.chao1	20.247440	12.961481	2.501562	28.17833	13.741182
S.ACE	193.949540	117.424538	72.533352	165.95257	117.889902
se.ACE	7.411584	5.482515	4.106573	7.25455	5.261013

Fig. 3 **a** Relative abundance of OTUs (core OTUs) in Drass (DR), Antarctic 1 (ANT1), Antarctic 1 (ANT2), Arctic 1 (ARC1) and Arctic 2 (ARC2). **b** Relative abundance of OTUs (unique OTUs and rare OTUs) in Drass (DR), Antarctic 1 (Ant1), Antarctic 1 (Ant2), Arctic 1 (Arc1) and Arctic 2 (Arc2)



Actinobacteria form the core phyla in cold deserts under investigation, and being mycelia formers, they are adapted to oligotrophic environments where the hyphae allow the bacteria to retrieve water and nutrients via pores in soil (Keki et al. 2013). Lesser representation of *Bacteroidetes* and *Cyanobacteria* in the soil of Drass supports the fact that Drass soil is relatively more oligotrophic than the Antarctic and Arctic soils. *Bacteroidetes* are positively correlated with the organic carbon content of the soil (Fierer et al. 2007). These are typically decomposers and commonly associated with substrates rich in organic carbon contributing to the turnover of polymeric carbon (Fazi et al. 2005; Aislabie et al. 2009; Zakhia et al. 2008) in the soil. *Cyanobacteria* are the predominant biota in cold polar environments and play an important role in the carbon and nitrogen economy of tundra and polar desert soils (Bonilla et al. 2005; Adams et al. 2006; Vincent 2007; Varin et al. 2012). This photosynthetic phylum is either reported to be present in low frequency or absent in the Himalayan regions (Pradhan et al. 2010; Shivaji et al. 2011; Stres et al. 2014). *Gemmatimonadetes* and *Verrucomicrobia* found in the present study were reported from other cold habitats, though at a low frequency (Steven et al. 2007; Wagner et al. 2009; Pradhan et al. 2010; Wu et al. 2012). The core phyla obtained in the present study have also been reported from other cold habitats. *Acidobacteria* have been reported from the Tibetan plateau glacier, China, Mount Everest, Nepal, John Evans glacier, Canada Bench Glacier, Alaska, Schirmacher Oasis soil and Antarctic Siberian tundra soil samples (Zhou et al. 1997, Shivaji et al. 2004; Skidmore et al. 2005; Cheng and Foght 2007; Liu et al. 2009; Campbell et al. 2010; Wu et al. 2012; Kim et al. 2014). *Proteobacteria* have been reported to be the dominant in the cold soils of the Himalayan mountains (Pradhan et al. 2010; Shivaji et al. 2011, Srinivas et al. 2011); Malan ice core drilled from the Tibetan Plateau (Xiang et al. 2004); surface sediment from the Arctic Ocean (Xuezheng et al. 2014); High-Arctic snow over sea ice (Moller et al. 2013) and Antarctic samples (Tytgat et al. 2014). The dominant Order *Myxococcales* of *Deltaproteobacteria* detected in Drass have been reported in Siberia, Arctic. *Bacteroidetes* has been reported as one of the abundant phyla from cold habitats (Wu et al. 2012; Moller et al. 2013). *Actinobacteria* reported to be the abundant phyla in cold habitats (Wu et al. 2012; Tytgat et al. 2014) and predominant phyla in Rookkund glacier, Himalayas (Pradhan et al. 2010).

The presence of *Firmicutes* in other cold habitats (Liu et al. 2009; Wagner et al. 2009) is well documented in the literature. Earlier studies on Himalayas (Pradhan et al. 2010; Shivaji et al. 2011) reported *Firmicutes* as the abundant phylum contrary to the present study. Negligible reads detected in Arctic subsamples is in accordance with Chu et al. (2010) who classified *Firmicutes* as the rarer phyla

in Arctic soil. *Armatimonadetes*, *Chloroflexi*, *Nitrospirae*, candidate phylum *TM7* and *Planctomycetes* are present in Drass soil at a low frequency as in other cold habitats (Shivaji et al. 2004; Cheng and Foght 2007; Wu et al. 2007; Li et al. 2008; Liu et al. 2009; Wagner et al. 2009; Pradhan et al. 2010; Shivaji et al. 2011; Stres et al. 2014). *WS3* was detected in Drass soil only and completely absent in Antarctic and Arctic samples although few reports that justify the presence of Phylum *WS3* in Antarctic and Arctic (Schutte et al. 2010; Tytgat et al. 2014) also. *Tenericutes* detected in Antarctic samples only in the present study have also been reported from few proglacial soils within the Himalayan regions (Srinivas et al. 2011; Stres et al. 2014).

Conclusion

On analyzing, and comparing the bacterial diversity of soil metagenome of Drass with Antarctic and Arctic metagenome, it was found that though most of the bacterial phyla present was common, but only the pattern of their dominance differed. Phylum *Acidobacteria* were dominant in the Drass soil, phylum *Firmicutes* were dominant in the Antarctic (not detected in Arctic soil), and *Proteobacteria* were dominant in the Arctic soil. Comparative sequence analysis also suggests that Drass soil is more arid than the Arctic and Antarctic soils and further resembles more to the Arctic than Antarctic soil or other Himalayan regions. Present study compliments the scientific reports that suggest that the bacterial community of any niche is specific, qualitatively and quantitatively designed by the combination of factors such as temperature, pH, salinity, water content, nutrients and carbon content.

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