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Biodiversity and Biotechnological Applications of Psychrotrophic Microbes Isolated from Indian Himalayan Regions

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COLUMN ARTICLE

Abstract

Extreme cold environments are the hot spots of biodiversity of diverse groups of microbes including archaea, bacteria and fungi. Prospecting the cold habitats of the Indian Himalayan region has led to the isolation of a great diversity of psychrotrophic microbes. The cold-adapted microbes have potential biotechnological applications in agriculture, medicine and industry as they can produce cold-adapted enzymes, anti-freezing compounds, antibiotics and possess diverse multifarious plant growth promoting attributes. Cold adapted microbes are ubiquitous in nature and can be isolated from permanently ice-covered lakes, cloud glaciers, and hilly regions. Microbes recovered using culture dependent techniques belong to different phylum Actinobacteria, Bacteroidetes, Basidiomycota, Chlamydiae, Chloroflexi, Cyanobacteria, Firmicutes and Proteobacteria of diverse genera namely: *Arthrobacter, Bacillus, Brevundimonas, Cellulosimicrobium, Citricoccus, Enterobacter, Exiguobacterium, Flavobacterium, Janthinobacterium, Lysinibacillus, Methylobacterium, Paenibacillus, Pantoea, Planococcus, Pontibacillus, Providencia, Pseudomonas, Psychrobacter, Rhodococcus, Sanguibacter, Sphingobacterium, Sporosarcina, Staphylococcus, Stenotrophomonas* and *Virgibacillus.* The cold adapted microbes with multifunctional attributes may be applied in industry and agriculture sectors.

Keywords: Biodiversity; Cold Desert; Indian Himalayas; Psychrotrophic Microbes; Sub-Glacial Lakes

INTRODUCTION

The microbiome of cold habitat is of particular importance in global ecology since the majority of terrestrial and aquatic ecosystems of our planet are permanently or seasonally submitted to cold temperatures. The psychrotrophic microbes have been isolated from cold environments including permafrost soils, glaciers, sub-glacial lakes and hilly area. Microbial communities from cold habitat have attained the focus of applied research not only in terms of biotechnological prospects but also to understand the use of primitive analogues of biomolecules existed during early Earth environments [1,2]. The microbiomes of cold environments of Indian Himalayan regions have been extensively investigated in the past few years with a focus on culture dependent [3-8]. Many novel microbes have been sort out from cold environments worldwide including *Sphingobacterium antarcticus*, *Psychromonas ingrahamii*, *Exiguobacterium soli*, *Cryobacterium roopkundense*, *Sphingomonas glacialis*, *Pedobacter arcticus*, *Sphingobacteri-*

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um psychroaquaticum, *Lacinutrix jangbogonensis*, *Massilia eurypsychrophila*, *Glaciimonas frigoris* and *Psychrobacter pocilloporae* [9-14]. Along with novel species of psychrotrophic microbes, some microbial species including *Arthrobacter nicotianae, Brevundimonas terrae, Paenibacillus tylopili* and *Pseudomonas cedrina* have been reported first time from cold deserts of NW Himalayas and exhibited multifunctional plant growth promoting (PGP) attributes at low temperatures [5]. The psychrotrophic microbial species *Aurantimonas altamirensis, Bacillus baekryungensis, B. marisflavi, Desemzia incerta, Paenibacillus xylanexedens, Pontibacillus* sp., *Providencia* sp., *Pseudomonas frederiksbergensis, Sinobaca beijingensis* and *Vibrio metschnikovii* have been reported first time from high altitude and low temperature environments of Indian Himalayas [6], where as in another study on wheat microbiome in diverse agro-ecological ecosystem in India, the two psychrotrophic bacteria namely *Arthrobacter methylotrophus* and *Pseudomonas rhodesiae* have been sort out first time [3]. The psychrophilic and psychrotrophic *Bacillus* and *Bacillus* derived genera such as *Bacillus altitudinis, B. amyloliquefaciens, B. muralis, B. psychrosaccharolyticus, Paenibacillus lautus, Paenibacillus pabuli*, *Paenibacillus terrae* and *Paenibacillus tylopili*, with efficient plant growth promoting (PGP) attributes have been reported first time by Yadav., *et al.* [15].

The diversity of microorganisms inhabiting cold environments has been extensively investigated in the past few years with a focus on culture dependent techniques. The different groups of microbes have been reported, which included different phylum mainly phylum Actinobacteria, Bacteroidetes, Basidiomycota, Chlamydiae, Chloroflexi, Cyanobacteria, Firmicutes and Proteobacteria. Overall the distribution of psychrotrophic microbes varied in all bacterial phyla, Proteobacteria were most dominant followed by firmicutes and actinobacteria. Least number of microbes was reported from phylum Chlamydiae followed by Chloroflexi. On review of different cold environments in Indian Himalayan regions (Permanently ice-covered lakes, ice caped rivers and glaciers), it was found that 8 different phylum have been sort out belonging to different phylum included as Proteobacteria (42.57%), Firmicutes (32.94%), Actinobacteria (17.78%), Bacteroidetes (2.62%), Basidiomycota (1.75%), Cyanobacteria (1.17%), Chlamydiae (0.58%) and Chloroflexi (0.58%) (Figure 1). On review of fifteen different extreme cold environments of Indian Himalayan regions including, Glacier [Roopkund glacier, Pindari glacier, Gangotri glacier, Lahaul and Spiti]; Sub-glacial lakes [Chandratal Lake, Dal Lake, Dashair Lake, Gurudongmar lake, Pangong Lake]; Cold desert of Himalayas [Chumathang, Khardungla Pass, Rohtang Pass]; Ice-coped revivers [Indus River, Zanskar River, Beas River]. On study of microbiome of cold environments from Indian Himalayan regions, it was found that, more than 66 different genera of 8 different phyla have been sort out and characterized for different biotechnological prospective. There were many psychrotrophic bacteria have been reported as niche specific as well as common bacteria from diverse site survey e.g. among 66 different genera, *Arthrobacter, Bacillus, Exiguobacterium, Paenibacillus, Planococcus, Pseudomonas, Psychrobacter, Providencia, Sporosarcina* and *Staphylococcus* were ubiquitous in nature and have been reported all fifteen sites, whereas some unique microbes as niche-specific such as *Stenotrophomonas* from Indus river, *Klebsiella, Sinobaca* and *Vibrio* from Zanskar river, *Aurantimonas* from Beas River, *Desemzia* from Pangong Lake, *Variovorax* from Dashair Lake, *Duganella, Duganella, Herbaspirillum, Iodobacter, Novosphingobium, Oxalobacteraceae, Paucibacter, Pectobacterium, Pseudoalteromonas, Pseudomonadaceae* and *Undibacterium* from Gurudongmar Lake, *Microbacterium and Aeromonas from* Chandratal *Lake, Agromyces, Brevibacterium, Cedecea, Enterobacter, Erwinia* and *Micrococcus* from Dal Lake, *Mortierella* from Khardungla Pass, *Paracoccus* from Chumathang, *Methylobacterium* from Rohtang Pass, *Citricoccus* from Roopkund glacier, *Acidovorax* from Pindari glacier, *Brevibacillus* from Gangotri glacier and *Rahnella* from Lahaul and Spiti have been reported from the single site survey [1,2,5-8,15-32] (Figure 2).

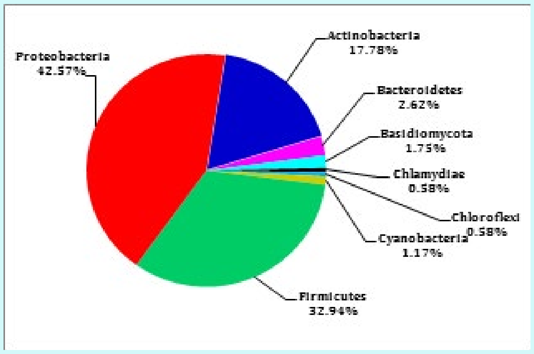

Figure 1: Distribution of different phylum of psychrotrophic microbes isolated from Indian Himalayan regions.

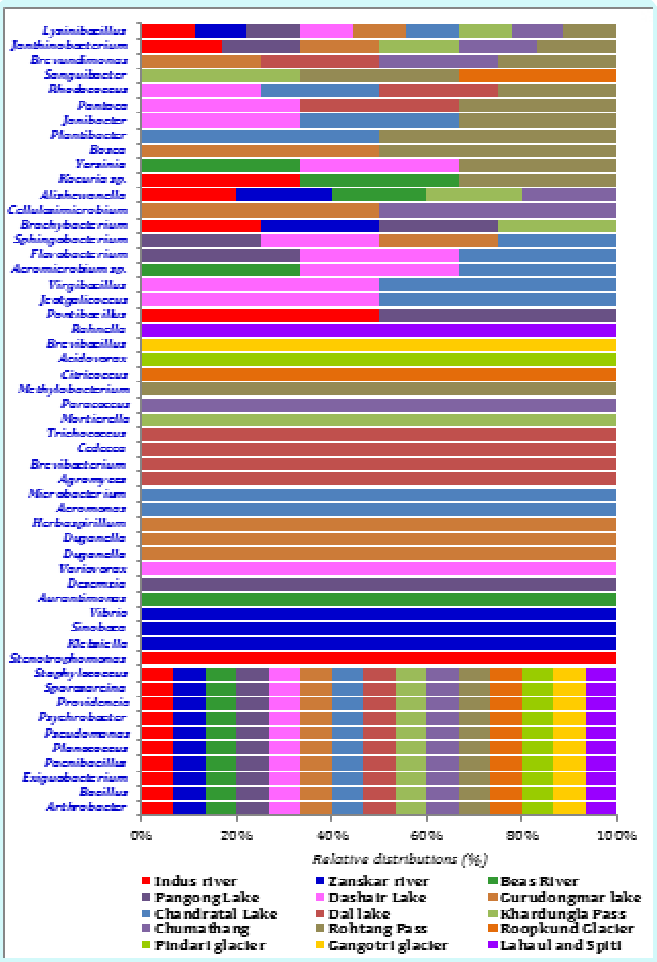

Figure 2: Relative distribution of different genera, isolated from cold environments of Indian Himalayan Regions.

Based on a comprehensive literature analysis psychrotrophic members of phylum actinobacteria has been reported from different genera such as *Acidimicrobium, Actinomyces, Arthrobacter, Bifidobacterium, Cellulomonas, Clavibacter, Corynebacterium, Frankia, Microbacterium, Micrococcus, Mycobacterium, Nocardia, Propionibacterium, Pseudonocardia, Rhodococcus, Sanguibacter* and *Streptomyces.* The member of *Arthrobacter* species were most dominant actinobacteria isolated from cold environments which included *Arthrobacter antarcticus, A. aurescens, A. chlorophenolicus, A. koreensis, A. methylotrophus, A. nicotianae, A. nicotinovorans, A. nitroguajacolicus, A. polychromogenes, A. psychrochitiniphilus, A. psychrolactophilus, A. sulfonivorans, A. sulfurous,* and *A. ureafaciens* [3-8,28,33,34].

Many *Bacillus* and *Bacillus* derived genera (BBDG) have been reported from cold environments such as *Bacillus aerius, Bacillus aerophilus, Bacillus stratosphericus, Bacillus altitudinis* [35], *Bacillus thuringiensis* [36], *Bacillus megaterium* [37], *Paenibacillus glacialis* [38] and *Bacillus cereus*, *Bacillus amyloliquefaciens, Bacillus cibi, Bacillus pumilus, Lysinibacillus fusiformis* [39,40]. Cold adapted Bacilli have been reported from different site survey in Indian Himalayan regions e.g. *Bacillus acidicola, B. altitudinis, B. amyloliquefaciens, B. aryabhattai, B. baekryungensis, B. cereus, B. cibi, B. circulans, B. firmus, B. flexus, B. licheniformis, B. marisflavi, B. megaterium, B. mojavensis, B. muralis, B. psychrosaccharolyticus, B. pumilus, B. simplex, B. sonorensis, B. subtilis, B. thuringiensis* and *B. weihenstephanensis.* Among in phylum Firmicutes, other *Bacillus* derived genera were *Exiguobacterium mexicanum, E. acetylicum, E. antarcticum, E. artemiae, E. aurantiacum, E. homiense, E. indicum, E. marinum, E. sibiricum* and *E. soli; Paenibacillus amylolyticus, P. lautus, P. pabuli, P. terrae, P. tylopili* and *P. xylanexedens; Planococcus antarcticus, P. donghaensis* and *P. kocurii* [3-8,25,28,33,34,41,42]. The phylum proteobacteria are a major group of Gram-negative bacteria which included α/β/γ proteobacteria which have been reported as psychrophilic as well as psychrotolerant e.g. *Pseudomonas aeruginosa, P. antarctica, P. azotoformans, P. baetica, P. cedrina, P. corrugate, P. costantinii, P. deceptionensis, P. extremaustralis, P. extremorientalis, P. fluorescens, P. fragi, P. frederiksbergensis, P. geniculata, P. gessardii, P. graminis, P. jessani, P. kilonensis,* *P. koreensis, P. lurida, P. mediterranea, P. moraviensis, P. orientalis, P. pavonaceae, P. peli, P. plecoglossicida , P. poae , P. psychrophila, P. putida, P. reactans, P. rhodesiae, P. simiae, P. stutzeri, P. syringae, P. teessidea, P. tolaasii, P. trivialis, P. vancouverensis* and *P. xanthomarina* [3,8,28,29,34,41,42].

Prospecting the cold habitats has led to the isolation of a great diversity of psychrotrophic microorganisms. The cold-adapted microbes have potential biotechnological applications in agriculture, medicine and industry. The microbial diversity from the cold environment could serve as a database for selection of bio-inoculants with PGP ability and could be used for improving the growth and yield of crops grown at high altitudes with prevailing low temperatures [3,5,6,8,43]. The psychrophilic/psychrotrophic/psychrotolerant microbes from Himalayan cold desert, glaciers, ice-coped revivers and different sub-glacial lakes could be used in agriculture, medicine and industry as they can produce cold-adapted enzymes (Amylase, cellulase, chitinase, laccase, lipase, pectinase, protease, xylanase, β-galactosidase and β-glucosidase), anti-freezing compounds, antibiotics and possesses diverse multifunction plant growth promoting attributes (production of ammonia, hydrogen cyanide, indole-3-acetic acid, and siderophore; solubilization of phosphorus, potassium and zinc; 1-aminocyclopropane-1-carboxylate deaminase activity and biocontrol activity against plant pathogenic microbes. Psychrotrophic plant growth promoting (PGP) microbes have been shown to promote plant growth either directly by biological N_2 fixation; solubilization of minerals such as phosphorus, potassium and zinc; production of siderophores and plant growth hormones or indirectly, via production of antagonistic substances by inducing resistance against plant pathogens under the native conditions from they have been isolated [3,4,15]. The psychrotrophic PGP microbes (*Arthrobacter, Bacillus, Brevundimonas Burkholderia, Pseudomonas, Citricoccus, Exiguobacterium, Flavobacterium, Janthinobacterium, Kocuria, Lysinibacillus, Methylobacterium, Microbacterium, Paenibacillus, Providencia* and *Serratia*) can have an impact on plant growth providing the plant with compound(s) of microbial origin for facilitating the uptake of nutrients from the environment [3,5,28,44,45]. These psychrotrophic microbes have potential benefits on the growth and yield of crops. The environmental health and soil productivity have been affected by use of psychrotropic microbes worldwide.

Enzymes from psychrophiles have become interesting for industrial applications, partly because of ongoing efforts to decrease energy consumption. These enzymes provide opportunities to study the adaptation of life to low temperature and the potential for biotechnological exploitation [2,31]. Most of the work that has been conducted on psychrophilic microbes focused on cold-adapted enzymes such as amylase, protease, lipase, pectinase, xylanase, cellulase, β-galactosidase, β-glucosidase and chitinase [31]. Many psychrotrophic microbes *Arthrobacter*, *Bacillus*, *Flavobacterium*, *Marinomonas*, *Moraxella*, *Paenibacillus*, *Planococcus*, *Pseudomonas*, *Psychrobacter*, *Shewanella* and *Vibrio* produced cold-actives extracellular hydrolytic enzymes with potential application in agriculture, medicine and industry [5,6,8,28,31]. Psychrophilic microbes (*Eupenicillium crustaceum*, *Paceliomyces* sp., *Bacillus atropheus* and *Bacillus* sp.) can be applied for biodegradation of agro wastes at low temperatures [46]. The psychrophilic microbes have been potential application in cryosurgery as they produced anti-freezing compounds at low temperatures [28]. Anti-freezing compounds are useful in cryosurgery and also in the cryopreservation of whole organisms, isolated organs, cell lines and tissues. In food industry, anti-freezing proteins (AFPs) can be used to improve the quality of frozen food. Improved cold tolerance in fishes has been achieved in some cases by direct injection of AFPs and in another case by transgenic expression of an AFP.

CONCLUSION AND FUTURE VISION

The psychrophilic and psychrotolerant microbes from Indian Himalayan regions have engrossed in the scientific community due to having the potential beneficial role in PGP under the low temperature conditions and produced industrially important cold-active extracellular hydrolytic enzymes which could be applied in a broad range of industrial, agricultural and medical processes. Psychrotrophic microbes could be valuable in agriculture as bio-inoculants and biocontrol agents for low temperature habitats. The use of psychrophiles as biofertilizers, biocontrol agent and bioremediators would be of great use in agriculture under cold climatic conditions. The comprehensive analyses of diversity of different genera by prospecting extreme cold environments helped in the development of a huge database including baseline information on the distribution of psychrotrophic microbes in different niches and identifying niche-specific microbes. This database also helped in identifying novel microbes with plant growth promoting attributes and biomolecules. The plant growth promotion potential of the microbial strain could be used as a bioinoculants in the hill and mountain agro ecosystems, where temperature is a major determinant of plant and microbial activity. The selection of native functional plant growth promoting microorganisms is a mandatory step for reducing the use of energy intensive chemical fertilisers. The cultures tolerant to low temperatures represent a rich bioresources for useful genes and alleles, which can aid in the generation of abiotic cold-tolerant transgenics.

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COMPETING INTERESTS

The authors declare no conflict of interest.

BIBLIOGRAPHY

1. Yadav AN. "Bacterial diversity of cold deserts and mining of genes for low temperature tolerance". Ph.D. thesis IARI, New Delhi/BIT, Ranchi (2015): 234.

2. Saxena AK., *et al*. "Microbial diversity of extreme regions: An unseen heritage and wealth". *Indian Journal of Plant Genetic Resource* 29.3 (2016): 246-248.

3. Verma P., *et al*. "Assessment of genetic diversity and plant growth promoting attributes of psychrotolerant bacteria allied with wheat (Triticum aestivum) from the northern hills zone of India". *Annals of Microbiology* 65.4 (2015): 1885-1899.

4. Verma P., *et al*. "Molecular diversity and multifarious plant growth promoting attributes of Bacilli associated with wheat (Triticum aestivum L.) rhizosphere from six diverse agro-ecological zones of India". *Journal of Basic Microbiology* 56.1 (2016): 44-58.

5. Yadav AN., *et al*. "Prospecting cold deserts of north western Himalayas for microbial diversity and plant growth promoting attributes". *Journal of Bioscience and Bioenginerering* 119.6 (2015): 683-693.

6. Yadav AN., *et al*. "Culturable diversity and functional annotation of psychrotrophic bacteria from cold desert of Leh Ladakh (India)". *World Journal of Microbiology and Biotechnology* 31.1 (2015): 95-108.

7. Yadav AN., *et al*. "Diversity and phylogenetic profiling of niche-specific Bacilli from extreme environments of India". *Annals of Microbiology* 65.2 (2015): 611-629.

8. Yadav AN., *et al*. "Extreme Cold Environments: A Suitable Niche for Selection of Novel Psychrotrophic Microbes for Biotechnological Applications". *Advance in Biotechnology and Microbiology* 2.2 (2017): 1-4.

9. Yadav AN., *et al*. "Psychrotrophic microbes: Diversity analysis and bioprospecting for industry and agriculture". In: 85th Annual Session of NASI and the Symposium on "Marine and Fresh Water Ecosystems for National Development" (2015).

10. Chaturvedi P., *et al*. "Exiguobacterium soli sp. nov., a psychrophilic bacterium from the McMurdo Dry Valleys, Antarctica". *International Journal of Systematic and Evolutionary Microbiology* 58.10 (2008): 2447-2453.

11. Reddy G., *et al*. "Cryobacterium roopkundense sp. nov., a psychrophilic bacterium isolated from glacial soil". *International Journal of Systematic and Evolutionary Microbiology* 60.4 (2010): 866-870.

12. Shivaji S., *et al*. "Sphingobacterium antarcticus sp. nov., a psychrotrophic bacterium from the soils of Schirmacher Oasis, Antarctica". *International Journal of Systematic and Evolutionary Microbiology* 42.1 (1992): 102-106.

13. Zachariah S., *et al*. "Psychrobacter pocilloporae sp. nov., isolated from a coral, Pocillopora eydouxi". *International Journal of Systematic and Evolutionary Microbiology* 66.12 (2017): 5091-5098.

14. Yadav AN., *et al*. "Microbiome of Indian Himalayan regions: Molecular diversity, phylogenetic profiling and biotechnological applications". In: Proceeding of 86th Annual Session of NASI and Symposium on "Science, Technology and Entrepreneurship for Human Welfare in The Himalayan Region" (2016): 83.

15. Yadav AN., *et al*. "Bioprospecting of plant growth promoting psychrotrophic Bacilli from cold desert of north western Indian Himalayas". *Indian Journal Experimental Biology* 54.2 (2016): 142-150.

16. Yadav AN., *et al*. "Diversity and Phylogeny of low temperature plant growth promoting fluorescent Pseudomonas isolated from cold desert of north western Himalayas". In: Proceeding of 54th AMI, International symposium on "Frontier Discoveries and Innovations in Microbiology and its Interdisciplinary Relevance". (2013): 396-397.

17. Yadav AN., *et al*. "Cold tolerant microbes from Indian Himalayas: diversity and potential applications in biotechnology and agriculture". In: Proceeding of 84th Annual Session of NASI and Symposium on "Desert Science-Opportunities and challenges" (2014): 17.

18. Baghel V., *et al*. "Psychrotrophic proteolytic bacteria from cold environment of Gangotri glacier, Western Himalaya, India". *Enzyme and Microbial Technology* 36.5 (2005): 654-659.

19. Gulati A., *et al*. "Characterization of phosphate-solubilizing fluorescent pseudomonads from the rhizosphere of seabuckthorn growing in the cold deserts of Himalayas". *Current Microbiology* 56.1 (2008): 73-79.

20. Pradhan S., *et al*. "Bacterial biodiversity from Roopkund glacier, Himalayan mountain ranges, India". *Extremophiles* 14.4 (2010): 377-395.

21. Vyas P., *et al*. "Cold-adapted and rhizosphere-competent strain of Rahnella sp. with broad-spectrum plant growth-promotion potential". *Journal of Microbiology and Biotechnology* 20.12 (2010): 1724-1734.

22. Shivaji S., *et al*. "Bacterial diversity of soil in the vicinity of Pindari glacier, Himalayan mountain ranges, India, using culturable bacteria and soil 16S rRNA gene clones". *Extremophiles* 15.1 (2011): 1-22.

23. Anupama P., *et al*. "A psychrophilic and halotolerant strain of Thelebolus microsporus from Pangong Lake, Himalaya". *Mycosphere* 2.5 (2011): 601-609.

24. Srinivas TNR., *et al*. "Comparison of bacterial diversity in proglacial soil from Kafni Glacier, Himalayan Mountain ranges, India, with the bacterial diversity of other glaciers in the world". *Extremophiles* 15.6 (2011): 673-690.

25. Pandey S., *et al*. "Phylogenetic diversity and characterization of novel and efficient cellulase producing bacterial isolates from various extreme environments". *Bioscience, Biotechnology, Biochemistry* 77.7 (2013): 1474-1480.

26. Sahay H., *et al*. "Cold-active hydrolases producing bacteria from two different sub-glacial Himalayan lakes". *Journal of Basic Microbiology* 53.8 (2013): 703-714.

27. Saxena AK., *et al*. "Use of Microbes from extreme environments for the benefits of agriculture". In: Afro-Asian Congress on microbes for human and environmental health (2014).

28. Singh RN., *et al*. "First, High quality draft genome sequence of a plant growth promoting and Cold Active Enzymes producing psychrotrophic Arthrobacter agilis strain L77". *Standand in Genomic Science* 11 (2016): 54.

29. Srivastava AK., *et al*. "Diversity analysis of Bacillus and other predominant genera in extreme environments and its utilization in Agriculture" (2014).

30. Venkatachalam S., *et al*. "Culturable and Culture-Independent Bacterial Diversity and the Prevalence of Cold-Adapted Enzymes from the Himalayan Mountain Ranges of India and Nepal". *Microbial Ecology* 69.3 (2015): 472-491.

31. Yadav AN., *et al*. "Cold active hydrolytic enzymes production by psychrotrophic Bacilli isolated from three sub-glacial

lakes of NW Indian Himalayas". *Journal of Basic Microbiology* 56.3 (2016): 294-307.

32. Yadav AN., *et al*. "Current applications and future prospects of eco-friendly microbes". *EU Voice* 3.1 (2017):1-3.
33. Rana KL., *et al*. "Biotechnological applications of endophytic microbes associated with barley (Hordeum vulgare L.) growing in Indian Himalayan regions". In: Proceeding of 86th Annual Session of NASI & Symposium on "Science, Technology and Entrepreneurship for Human Welfare in The Himalayan Region" (2016): 80.
34. Rana KL., *et al*. "Diversity and biotechnological applications of endophytic microbes associated with maize (Zea mays L.) growing in Indian Himalayan regions". In: Proceeding of National Conference on Advances in Food Science and Technology (2017): 41-42.
35. Shivaji S., *et al*. "Bacillus aerius sp. nov., Bacillus aerophilus sp. nov., Bacillus stratosphericus sp. nov. and Bacillus altitudinis sp. nov., isolated from cryogenic tubes used for collecting air samples from high altitudes". *International Journal of Systematic and Evolutionary Microbiology* 56.7 (2006): 1465-1473.
36. Das J., *et al*. "Microbial population dynamics, especially stress tolerant Bacillus thuringiensis, in partially anaerobic rice field soils during post-harvest period of the Himalayan, island, brackish water and coastal habitats of India". *World Journal of Microbiology and Biotechnology* 24.8 (2008): 1403-1410.
37. Trivedi P., *et al*. "Plant growth promotion abilities and formulation of Bacillus megaterium strain B 388 (MTCC6521) isolated from a temperate Himalayan location". *Indian Journal of Microbiology* 48.3 (2008): 342-347.
38. Kishore KH., *et al*., "Paenibacillus glacialis sp. nov., isolated from the Kafni glacier of the Himalayas, India". *International Journal of Systematic and Evolutionary Microbiology* 60.8 (2010): 1909-1913.
39. Sahay H., *et al*. "Hot springs of Indian Himalayas: Potential sources of microbial diversity and thermostable hydrolytic enzymes". *3 Biotech* 7.2 (2017): 118.
40. Verma P., *et al*. "Hydrolytic enzymes production by thermotolerant Bacillus altitudinis IARI-MB-9 and Gulbenkiania mobilis IARI-MB-18 isolated from Manikaran hot springs". *International Journal of Advanced Research* 3.9 (2015): 1241-1250.
41. Saxena AK., *et al*. "Biotechnological applications of microbes isolated from cold environments in agriculture and allied sectors". In: International Conference on "Low Temperature Science and Biotechnological Advances" (2015).
42. Kumar V., *et al*. "β-Propeller phytases: Diversity, catalytic attributes, current developments and potential biotechnological applications". *International Journal of Biological Macromolecules* 98 (2017): 595-609.
43. Suman A., *et al*., "Endophytic Microbes in Crops: Diversity and Beneficial impact for Sustainable Agriculture". In: Singh., *et al*. (eds), Microbial Inoculants in Sustainable Agricultural Productivity, Research Perspectives. Springer-Verlag, India (2016): 117-143.
44. Verma P., *et al*. "Alleviation of cold stress in wheat seedlings by Bacillus amyloliquefaciens IARI-HHS2-30, an endophytic psychrotolerant K-solubilizing bacterium from NW Indian Himalayas". *National Journal of Life Sciences* 12.2 (2015): 105-110.
45. Yadav AN., *et al*. "Mitigation of cold stress for growth and yield of wheat (Triticum aestivum L.) by psychrotrophic pseudomonads from cold deserts of Indian Himalayas" In: Proceeding of 56th AMI 20105 and International symposium on "Emerging Discoveries in Microbiology" (2015).
46. Shukla L., *et al*. "Syntrophic microbial system for ex-situ degradation of paddy straw at low temperature under controlled and natural environment". *Journal of Applied Biology and Biotechnology* 4.2 (2016): 30-37.