



GEOMICROBIOLOGY: AN EMERGING SCIENCE AND WAY OUT TO ENVIRONMENTAL AND HEALTH PROBLEMS

Abirami Subramanian and Sushmitha Baskar*

Environmental Studies, School of Interdisciplinary and Transdisciplinary Studies (SOITS)
Indira Gandhi National Open University (IGNOU), Maidan Garhi, New Delhi 110068.

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Abstract

Geomicrobiology involves the role of microorganisms in geological environments and geochemical processes. Extreme environments such as caves, hot springs, deep-sea hydrothermal vents, marine environments, extreme cold environments such as Arctic and Antarctic, deserts, and heavily polluted environments, host diverse microbial species of varied interest. Researchers from various fields are partnering to investigate such microorganisms for a variety of environmental and health applications. It is estimated that our planet still has 99.99 percent of its microbial diversity unexplored. So, to delve deeply into the exploration of microbial diversity and its functions, the metagenomic approach acts as a lens for a broader picture than traditional culturing methods. The metagenomics approach entails the direct investigation of microorganisms in specific environmental samples. Several such discoveries have made significant contributions to medicine, pharmaceutical industries, bioremediation, biodegradation, biomineralization, biomining, biofuel production, space exploration, and other fields. Thus, this article discusses the identification of extreme microbes and their possible potential applications in the environment and health.

Keywords

Geomicrobiology, Extreme Environments.

INTRODUCTION

Geomicrobiology is a cutting-edge area of research that involves the interactions of microbes with earthly processes. Microorganisms though invisible are the rulers of the planet. Microorganisms isolated from extreme environments have a wide range of applications in environmental and health sciences. Our planet has room for every kind of extremities like boiling heat (Thermophiles) and freezing temperatures (Psychrophiles), highly acidic (Acidophiles) and basic conditions (Alkaliphiles), extremely high pressure (Barophiles) and low pressure, regions of high radiation intensities, nutrient-rich and deficient conditions, and also the combination of two or more such extremes. Microbes that live in harsh environments have a unique metabolic potential that allows them to survive in such extremities. Indeed, these microorganisms cannot survive in the absence of the extreme conditions to which they have adapted. To identify unknown microbial diversity conventional culture-dependent methods

are not enough, because these methods can be used only for the predefined hypothesis of known diversity. So to overcome the roadblock of conventional culture methods, the meta-analysis of the environmental samples i.e the culture-independent methods will pave the way. Recently the search for novel microbes in extreme environments has become the frontier research subject. To unravel the estimated 99.99 percentage of microbial diversity, metagenomics analysis will act as a lens to zoom in on the untapped organisms.

The main objective of this review paper is to discuss the identification and the applications of microorganisms isolated from extreme environments in the field of environment and healthcare.

Culture dependent and independent methods for characterization of geomicrobiome

The microbial diversity in environmental samples can be

extracted in two ways, as shown below.

- Culture dependent method
- Culture-independent method

Culture dependent methods

Various agar plate and liquid media methods are employed in the traditional approach to recover the microbiome from the intended environmental sample. Different microbial species require different compositions of medium for their growth. So, culture-dependent methods are commonly used for predefined microbial community targets. The typical culture

media should contain energy sources (Light, carbohydrates, energy from oxidation, etc.), sources of carbon, nitrogen, sulphur, and phosphorus, minerals like calcium (Ca^{2+}) magnesium (Mg^{2+}), and sodium (Na^+), vitamins, and growth factors (Amino acids, blood, and its derivatives, antioxidants, etc.). Different types of culture media used for the isolation of bacteria and fungi and their target species are depicted in Table 1. After the isolation of microbes via culture, the microbial communities can be characterized using either traditional biochemical methods or DNA extraction followed by sequencing.

Table 1: Culture media for bacteria and fungi with the target species.

Culture media for bacteria	Target species	Culture media for fungi	Target species
Nutrient agar	To isolate non-fastidious organisms like <i>klebsiella pneumonia</i> , <i>Morgnella morganni</i> , <i>Providencia alkalifaciens</i> , <i>Salmonella typhimurium</i>	Birdseed agar	<i>Cryptococcus neoformans</i>
Mac Conkey agar	<i>Enterobacteriaceae</i>	Brain-heart infusion agar	<i>Histoplasma capsulatum</i> and <i>Blastomyces dermatitidis</i>
Mannitol salt agar	<i>Staphylococcus aureus</i>	CHROMagar Candida medium	To isolate various <i>candida</i> species
Chocolate agar	To isolate pathogenic bacteria	Czapek-Dox agar	<i>Aspergillus</i> and <i>Penicillium</i> species.
Thayer-Martin agar	<i>Neisseria gonorrhoea</i>	Potato dextrose agar	To isolate most fungal species
Cetrimide agar	<i>Pseudomonas aeruginosa</i>	Potato flake agar	To isolate saprophytic and pathogenic fungi
Hektoen enteric agar	<i>Salmonella</i> sp. and <i>Shigella</i> sp.	Sabouraud dextrose agar	To isolate clinically important fungi

Culture-Independent methods

The steps involved in the culture independent method for the investigation of microbial diversity are given below (Simon and Daniel, 2011):

- Culture independent method involves the direct extraction of DNA and RNA from the environmental sample based on the need.
- In the case of DNA, the extracted DNA can be amplified for the construction of a metagenome library for bioprospecting application.
- To assess the taxonomic diversity, the extracted metagenomic DNA from the target sample is subjected to sequencing of phylogenetic marker genes.
- The extracted DNA can also be sequenced to assess the taxonomic diversity and their metabolic potential.

- To assess the microbial community functions and the active members of the microbial community the extracted RNA should be subjected to mRNA and rRNA analysis respectively.

Applications of Extremophiles in Environment and Health

The metabolic potential and the structural stability of the extremophiles have immense applications in environmental and health sectors. Scientific exploration of these resources is still in the infant stage of its development. Table 2 depicts the various applications of extremophiles.

Applications in Environment

Environmental health is the price we pay for all of the modern conveniences. Since the beginning of the green revolution and industrial revolution, the quality of earth's products such as air, water, soil, and other natural resources has begun to deteriorate. Thus it is our primary responsibility to restore

Table 2: Application of extremophiles in various environmental and health applications.

S. No.	Microbes	Habitat	Applications	Reference
1	Desmodesmus sp. MAS1	Soil and Lake water	Biofuel production	(Abinandan et al., 2018)
2	Heterochlorella sp. MAS3			
3	Nesterenkonia sp.	Hypersaline lake in Iran	Butanol, ethanol, and acetone production	(Amiri et al., 2016)
4	Pseudomonas fluorescens AH-40	Crudeoil polluted soil	Bioremediation of phenanthrene contaminated sites	(Mawad et al., 2020)
5	Streptomyces sps.	Bay of Bengal	The biosynthetic and bioactive compound	(Ghosh et al., 2020)
6	<i>Penicillium</i> sp. (GBPI_P155)	The soil of the Indian Himalayan region	Natural colour pigment (Orange)	(Pandey et al., 2018)
7	<i>Bacillus licheniformis</i>	Geothermal springs of Armenia and Nagorno Karabakh	Thermostable enzyme production	(Panosyan et al., 2020)
	<i>Anoxybacillus favithermus</i>			
	<i>Parageobacillus toebii</i>			
8	Pyrodictium delaneyi Su06	Deep-sea hydrothermal vent	Iron oxide reduction	(Kashyap and Holden, 2021)
9	Rhodothermus marinus	Hot spring	Biorefinery	(Kristjansdottir et al., 2020)
10	Geomyces sp.F09-T3-2	Marine sponges	Pectinolytic activity	(Poveda et al., 2018)
11	Penicillium glabrum SF-7123	Antarctic marine	Anti inflammatory and anti-neuroinflammatory	(Ha et al., 2020)
12	Haloferax volcani	Dead sea	Antioxidant, bioprocessing, bioremediation, carotenoid synthesis	(Haque et al., 2020)
13	Streptomyces sp. AMA50	Marine environment (Thailand)	Antifungal activity	(Sangkanu et al., 2021)
14	Serratiopeptidase (Isolated from Serratia marcescens)	The intestine of Bombyx mori L.	Anti-inflammatory, fibrinolytic effects	(Jadhav et al., 2020)
15	Archaeoglobus fulgidus	Hot springs, high-temperature oil fields	Biodegradation of Phthalic acid ester's (PAEs)	(Zhang et al., 2020)

environmental health by remediating and utilizing it sustainably. Fighting fateful harsh conditions like highly polluted environments, acid mine drainage, clearing landfills, restoring the degraded lands, and so on with the geologically extreme microbes have gained importance among environmental scientists. It is also of significant interest to employ them as an environmentally acceptable option for biofuel production, bioremediation, biodegradation, biomining, bioleaching, bioplastics, and for a variety of synthetic and toxic chemicals used in industrial applications.

Biofuel Production

Replacing fossil fuels with eco-friendly alternatives will be the greatest contribution to achieve sustainable development

goals (SDGs). The production of biofuels started with edible biomasses like sugarcane, wheat, maize, animal fats, and so on, and the second generation fuels gradually shifted to non-edible biomasses like waste, wood, straw, grasses, bagasse, etc. The third-generation biofuel production employs various microorganisms and microalgae as an efficient alternative to the previous generation of biofuels. To develop cost-effective and energy-efficient biofuels as a superior replacement for fossil fuels while simultaneously addressing rising energy demand is the need of an hour.

Enzymes

Extremophiles create extremozymes, which are stable enzymes that can resist extremes in temperature, pH, light intensity, and so on. Extremozymes have the potential

application in biotechnological advancements and in the case of biofuel production enzyme activity is an important process in the conversion of the raw biomass into biofuels. For instance, microalgae are extensively used for biofuel production as they can accumulate 50- 70% of lipids by CO₂ fixation (Saharan et al., 2013). The lipids can be further degraded by cellulose-degrading enzymes and thus producing bio-oils. Many acidophilic, halophilic, and thermophilic fungus and bacteria are also employed in the production of green energy generations.

Biomining

Biomining uses microorganisms to recover metals and ores from the environment, as well as to clean up metal-contaminated sites. The mining sites and mine drainages are well known for their acidic pH. The extreme acidity will degrade the lands and also the plant growth is inhibited thus causing land and resource degradation. Mining for the commercially important metals and ore in a sustainable way is the major challenge. Extremophiles will be the best solution to the challenge as they can resist a wide range of temperatures and pH fluctuations. Acidophilic microbes, for example, may keep their internal pH lower than other microbes, allowing them to endure such acidic environments without internal cell damage (Chen, 2021). Widely employed extremophiles in biomining include *Acidothiobacillus ferrooxidans*, *Pyrococcus furiosus*, *sulfolobus metallicus*, *Metalosphaera sedula*, etc.

Bioremediation

Microbes are known as potential remediators as they can degrade even complex chemical compounds and heavy metals. The bioremediation process though has many advantages it has some disadvantages such as the slow rate of degradation of pollutants. To overcome this, it is important to understand the microbial communities involved in the bioremediation process at the molecular level. Microbes that thrive in geological extremes are the ideal cleaners for man-made extremes. As they have the ability to adapt and respond to the increased contaminants in the surrounding environment. Extremophiles who live in hostile environments have developed mechanisms to mobilize, immobilize, convert, absorb, or biodegrade pollutants in an eco-friendly way. They also have the potential to transform toxic organic pollutants into nontoxic end products within a short period of time irrespective of their half-life (Dua et al., 2002; Reiger et al., 2002; Sinkkonen and Paasivirta, 2000). *Bacillus safensis*, *Haloferax mediterranei*, *Nitrosopumilus maritimus*, *Pseudomonas stutzeri*, *Halococcus salifodinae*, *Halobacterium noricense*, *methanotrix soehngeni*, *Methanococcus mazei*, *Halofera mediterranei*, *Rhodanobacter* sp. are some of the examples of extremophiles used in the field of bioremediation to remediate petroleum products, heavy metals, pesticides, wastewater and radionuclides (Kaushik et al., 2021).

Applications in Health

Modern medical science and technological advancements have cured a variety of previously incurable health conditions. Microorganisms that are resistant to contamination and fluctuation have a huge role in medicine. Extremophiles producing bioactive compounds, antimicrobial peptides, and enzymes are of great commercial interest and are considered a safe alternative for synthetic compounds.

Cure for Protein Misfolding Diseases

Proteins are highly susceptible to external and internal stress conditions. This stress may eventually lead to protein denaturation and paves way for various degenerative and neurodegenerative diseases like Alzheimer's disease, cystic fibrosis, Huntington's disease, Parkinson's disease. Recently, the stability of extremophiles and the extremozymes and extremolytes produced by them have gained importance in the field of regenerative medicine. Marine ecosystems serve as a reservoir for a variety of interesting neuroprotective compounds. In particular, Alzheimer's disease (AD) is a booming problem among the old-age population. Acetylcholinesterase (AChE) and Butyrylcholinesterase (BuChE) are considered as the important target enzymes to treat or prevent AD. The alkaloid compound derived from a marine bacterium *Rapidithrix thailandica* (Marinoquinoline A) (Sangnoi et al., 2008) and fungus *Acrostalagmus leteoalbus* TK-43 (Acrozines A-C) (Cao et al., 2019) are proved to have an inhibitory effect on AChE.

Cure for Antibiotic Resistance

The widespread use of antimicrobial compounds has caused the targeted microbial communities to adapt and resist the drugs. Therefore, antimicrobial/antibiotic resistance is the major concern face by health care sectors and remains a barrier to treat the diseases effectively. The development of new antimicrobials will be the breakthrough in the field of drug discovery. Since microorganisms have the capability to adapt and resist any kind of harsh conditions, the need for new antimicrobial compounds is in growing demand. Extremophiles present in the caves, marine environment, highly saline environment, and other extreme environments are the reservoir for the brandnew bioactive compounds that will be the promising key to break the challenge of antimicrobial resistance.

Peptides, diketopiperazines, and other bioactive compounds produced by extremophiles are of main interest for antimicrobial compounds. To name some microbes that are identified from extreme environments and are currently involved in the field of drug discovery are *Penicillium* sp. *Trichoderma velutinum*—(Singh et al., 2018), *Penicillium granulatum* MCCC 3A00475 (Niu et al., 2018), *Acremonium persicinum* SCSIO 115—(Luo et al., 2019), *Aspergillus sydowii* SP-1 (Li et al., 2017).

Other health care applications

The other medicinal applications of extremophiles include:

- The carotenoid pigment isolated from extreme halophilic archaeon *Halobacterium salinarum* is used as artificial retinas (Charlesworth and Burns, 2015).
- Radiation-resistant extremophiles can produce extremolytes and extremozymes which have the potential application as sunscreen, antioxidants, antiproliferative, anticancer agents, DNA repair mechanism, etc (Aguilera et al., 2002; Asgarani et al., 2000; Babu et al., 2015; Russo et al., 2008).
- Extremophiles are also used as alternative vaccine delivery vehicles.
- Health supplements
- Protein stabilizers
- Anti inflammatory and neuroinflammatory
- Skincare products
- Drug delivery

CONCLUSIONS

Geomicrobiological studies provide vast resources of microorganisms that can be explored for countless environmental and medicinal applications. Human beings are the sole responsible for the fateful extreme conditions like landfills, disposal of biomedical and electronic waste, polluted environments, radiation environments, land exploitation, and wastelands due to intensive agriculture practices and pesticide usage. As a result, climate change is the compliment we have gained for environmental destruction. Biomedical waste disposal is one of the major reasons for the development of antibiotic-resistant microbial strains. Therefore, understanding the microbes and their metabolism at the molecular level is the best of all solutions. Though the exploration of extremophiles and their applications in different fields have gained importance, it still needs a drift for active commercial applications. Thus, the metagenomics analysis of geomicrobiome will surely be the way out to the majority of environmental and health problems.

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