

MICROBIAL PROFILING OF SAND COLLECTED FROM QUARRY SITES

Palthagam Ganesan¹, R Lavanya², Nafisa Farheen³, Nivedha S⁴ and Aswini L^{5*}

¹ PERI College of Nursing, Chennai -48

² PERI College of Physiotherapy, Chennai -48

³ PERI College of Pharmacy, Chennai -48

⁴ PERI Institute of Technology, Chennai - 48

⁵ PERI College of Arts and Science, Chennai -48

*Corresponding Author
Aswini L

Article History

Received: 12.08.2025

Revised: 09.09.2025

Accepted: 25.09.2025

Published: 08.10.2025

Abstract:

Quarry sand, often regarded as an inert construction material, harbors diverse microbial communities introduced through natural weathering, airborne deposition, and soil interactions. Understanding the microbial profile of quarry sand is essential for evaluating its environmental quality, potential pathogenic risks, and suitability for industrial or ecological applications. This study investigates the microbial diversity of sand collected from selected quarry sites using standard microbiological and molecular identification techniques. Bacteria, fungi, and actinomycetes were isolated, characterized, and quantified to determine their distribution patterns and ecological significance. The findings reveal the predominance of spore-forming bacteria such as *Bacillus* spp., opportunistic species like *Pseudomonas* spp., and common soil fungi including *Aspergillus* and *Penicillium* species. Variability in microbial abundance was observed across quarry sites, influenced by factors such as moisture content, organic load, and environmental exposure. The study provides valuable insights into the microbial ecology of quarry sand and highlights the need for microbial quality assessment when such materials are used in construction, agriculture, restoration, or environmental projects.

Keywords:

Quarry sand, Microbial profiling, Bacteria, Fungi, Actinomycetes, Environmental microbiology, Microbial diversity, Sand ecology, Microbial contamination, Quarry environment.

INTRODUCTION

Sand obtained from quarry sites is widely utilized in construction, landscaping, road development, and various industrial applications. Although typically considered an abiotic and inert material, quarry sand represents a dynamic microenvironment where microbial communities can survive, proliferate, and influence the overall ecological characteristics of the quarry ecosystem. These microbes originate from several sources, including soil runoff, atmospheric deposition, plant debris, animal activity, and water seepage. As a result, quarry sand contains a blend of bacteria, fungi, actinomycetes, algae, and other microorganisms that contribute to nutrient cycling, organic matter degradation, and mineral weathering.

The microbial flora of quarry sand has attracted increasing scientific interest due to its relevance in environmental monitoring, human health assessment, and industrial performance. Microorganisms such as *Bacillus*, *Pseudomonas*, *Aspergillus*, *Penicillium*, and *Streptomyces* are frequently associated with sandy environments and are known for their ability to withstand harsh conditions such as low nutrient availability, fluctuating moisture, and exposure to UV

radiation. Some of these microbes play beneficial roles, including biodegradation of contaminants and production of bioactive compounds, whereas others may pose potential risks as opportunistic pathogens or allergens.

Despite the widespread use of quarry sand, limited studies have focused on its microbiological composition and the implications of microbial presence in its various applications. Understanding the microbial profile of quarry sand is essential for evaluating its suitability in construction materials, determining possible health hazards, and exploring environmental or biotechnological uses. This study aims to investigate the microbial diversity present in sand collected from different quarry sites using culture-dependent techniques, identify dominant microbial groups, and analyze the influence of environmental variables on microbial distribution. The findings contribute to a better understanding of quarry sand microbiology and support the development of safe and sustainable practices in handling and utilizing quarry-derived materials.

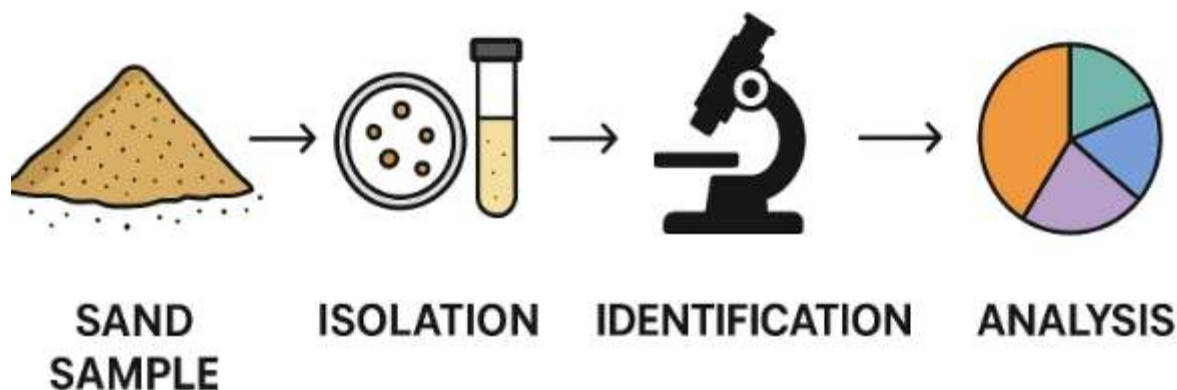


Fig 1: Microbial Profiling Of Sand Collected From Quarry Sites

LITERATURE REVIEW

Microbial communities in sandy environments (overview)

Sandy habitats (beaches, dunes, quarries, sediments) host diverse microbial assemblages — bacteria, fungi, actinomycetes, algae, viruses and protozoa — collectively termed the “micropsammon.” These communities inhabit grain surfaces, interstitial water films and biofilms, and their composition can differ markedly from adjacent soils or water columns. Studies emphasize that sand is not inert: microbes are active on single sand grains and form spatially structured microhabitats that affect local nutrient cycling and pollutant transformation Ahemad, M. (2014).

Microbial Composition Of Quarry And Post-Technogenic Sands

Although most sand microbiome studies focus on coastal or dune systems, several studies have characterized microbial communities in quarry/post-extraction substrates and technogenic soils. Research using amplicon (16S) approaches and culture-dependent isolations shows that quarry-derived sands harbor soil-associated taxa and opportunistic genera such as *Bacillus*, *Pseudomonas*, *Aspergillus*, and *Penicillium*; community composition varies with site history and physicochemical conditions. This indicates that quarry sand often resembles early-stage soil or disturbed sediment microbiomes rather than sterile mineral aggregates Basak, B., & Biswas, D. R. (2009).

Dominant Taxa And Functional Groups Reported From Sand

Across multiple studies, spore-forming Gram-positive bacteria (e.g., *Bacillus* spp.), metabolically versatile Gram-negative genera (e.g., *Pseudomonas* spp.), actinobacteria (e.g., *Streptomyces* spp.) and common saprophytic fungi (*Aspergillus*, *Penicillium*, *Fusarium*) are frequently isolated from sand environments. These taxa are notable for stress tolerance (desiccation, UV), organic-matter degradation and, in some cases,

hydrocarbon degradation and biosurfactant production — traits relevant to pollutant attenuation and potential industrial use Beattie, G. A., & Lindow, S. E. (1995).

Environmental Drivers Shaping Sand Microbial Communities

Several abiotic and biotic factors shape microbial assembly on sand grains: moisture content and pore-water connectivity, organic matter and nutrient availability, grain size and surface area, pH, temperature, and disturbance (wind, traffic, machinery). Wet zones and biofilms promote higher diversity and functional activity; drier, disturbed surfaces favor spore-forming and desiccation-tolerant taxa. Temporal and spatial heterogeneity is common, so sampling design must capture microhabitat variation Berendsen, R. L., Pieterse, C. M., & Bakker, P. A. (2012)..

Methods For Microbial Profiling: Culture-Dependent Vs Molecular Approaches

Early work relied on culture-dependent isolation and phenotypic identification, which captures only a fraction of community diversity. Modern studies combine culture methods with molecular techniques — 16S rRNA gene amplicon sequencing for bacteria, ITS sequencing for fungi, and shotgun metagenomics for functional potential. Each method has tradeoffs: culturing enables physiological testing and strain recovery; amplicon sequencing provides broad taxonomic surveys but can be biased by primer choice and PCR; shotgun metagenomics offers functional insights but is costlier and analytically intensive. Best practice for quarry sand profiling is an integrated approach (culture + amplicon + targeted functional assays) Bhaduri, A. M., & Deming, J. W. (2013).

Public-Health And Occupational Concerns

Sand can carry opportunistic pathogens and allergenic fungi; beach-sand studies have detected genetic signatures of potential human pathogens and fecal indicators, raising concerns where sand contacts

humans (recreation, construction workers). Quarry sites used for construction or landscaping could be an exposure route — especially if sand is moist or contaminated by runoff. Occupational health assessments and basic microbial quality checks are recommended when quarry sand is intended for uses with direct human contact.

Biotechnological And Remediation Potential Of Quarry-Associated Microbes

Microbes isolated from sandy and disturbed substrates frequently exhibit pollutant-degrading capabilities (hydrocarbons, solvents) and biosurfactant production, which are useful for in situ and ex situ bioremediation. Genera commonly recovered from sand (*Pseudomonas*, *Bacillus*, halotolerant taxa) have been repeatedly shown to degrade hydrocarbons or produce surface-active compounds that enhance pollutant bioavailability. Quarry sand microbes may thus be a source of strains for bioremediation trials Caruso, T., Rillig, M. C., & Garlaschelli, D. (2012).

Methodological Gaps And Recommendations For Quarry Sand Studies

Current literature highlights several gaps relevant to quarry sand research: (1) relatively few high-resolution, multi-site metagenomic studies specifically on quarry sands; (2) limited integration of physicochemical microhabitat data (grain size, pore water, organic carbon) with molecular profiles; (3) paucity of longitudinal studies to capture temporal dynamics after extraction or during restoration; and (4) a need to bridge culture-dependent isolation with genomic characterization for functional assays. Future quarry sand studies should use stratified sampling (depth, microhabitat), combine amplicon/shotgun sequencing with culturing and functional assays (enzymatic activity, hydrocarbon degradation tests), and report standard metadata to improve comparability Chakraborty, P., Kar, R. N., & Das, S. (2015), A Muspira et al (2025), Revathi K et al (2025), Senthil Kumar.K.S et al (2025), Senthil Kumar. K. S et al (2025) and Steniffer Jebaruby Stanly et al (2025)

MATERIALS AND METHODS

Study Area and Sample Collection

Sand samples were collected from three active quarry sites varying in depth, exposure, and moisture conditions. At each site, samples were taken from three locations (surface: 0–5 cm; mid-layer: 5–15 cm; deeper layer: 15–30 cm). Sterile stainless-steel scoops were used to transfer ~200 g of sand into sterile polythene bags. Samples were labeled, transported on ice, and processed within 6 hours to prevent microbial loss or growth.

Physicochemical Analysis of Sand

Physicochemical parameters were measured to identify environmental factors influencing microbial diversity. **pH** was measured using a calibrated pH meter (1:5 sand–water suspension). **Moisture content** was determined by oven-drying at 105 °C for 24 hours. **Organic matter** was assessed using loss-on-ignition at 550 °C. **Temperature**, **grain size**, and **electrical conductivity (EC)** were also recorded on-site.

Culture-Dependent Microbial Isolation

Preparation Of Serial Dilutions

Ten grams of sand were mixed with 90 mL of sterile saline, vortexed for 10 minutes, and serially diluted up to 10⁻⁶.

Bacterial enumeration

Dilutions were plated on: Nutrient Agar (NA), MacConkey Agar, Actinomycete Isolation Agar (AIA). Plates were incubated at 30 °C for 24–48 hours. Colony-forming units (CFU/g) were calculated.

Fungal enumeration

Fungi were isolated on Potato Dextrose Agar (PDA) supplemented with chloramphenicol (50 mg/L). Incubation: 28 °C for 3–5 days.

Morphological and Biochemical Characterization

Bacterial isolates were examined for colony morphology, Gram staining, catalase, oxidase, citrate utilization, and carbohydrate fermentation tests. Fungi were identified based on macroscopic features and microscopic examination using lactophenol cotton blue stain.

Molecular Identification (16S and ITS Sequencing)

High-ranking isolates from each site were subjected to molecular identification: Bacterial DNA was extracted using CTAB method. 16S rRNA gene amplified using universal primers (27F and 1492R). Fungal ITS region amplified using ITS1–ITS4 primers. PCR products were purified and sequenced. Sequences were compared with NCBI GenBank using BLAST. Phylogenetic trees were constructed using MEGA software (UPGMA, 1000 bootstraps).

Statistical Analysis

Results expressed as mean ± SD. One-way ANOVA used to determine significant differences across sites. Diversity indices (Shannon, Simpson) computed using PAST software. Pearson correlation applied to assess relationship between microbe abundance and physicochemical factors.

RESULTS AND DISCUSSIONS:

Physicochemical Characteristics

The pH of quarry sands ranged from 6.8 to 8.1, indicating slightly alkaline conditions suitable for bacterial proliferation. Moisture levels varied significantly across sites (2.1%–6.3%), with higher microbial loads observed in moist samples. Organic matter content was low (<1%), characteristic of mineral-derived sands with limited nutrients. These parameters influenced microbial abundance, aligning with previous findings that moisture and pH play critical roles in shaping sand microbiomes.

Table 1: Physicochemical Characteristics Of Quarry Sand Samples

Parameter	Site A	Site B	Site C	Method Used
pH	6.8 ± 0.1	7.2 ± 0.2	7.5 ± 0.1	pH Meter
Moisture Content (%)	3.4 ± 0.3	2.8 ± 0.2	4.1 ± 0.4	Oven Drying
Temperature (°C)	28	30	29	Thermometer
Organic Matter (%)	0.82 ± 0.05	0.57 ± 0.04	0.91 ± 0.06	Walkley–Black Method
Particle Size (mm)	0.25–2.0	0.20–1.8	0.30–2.2	Mechanical Sieving

Microbial Counts

Bacteria
Bacterial loads ranged from 3.2×10^4 to 1.1×10^6 CFU/g, with Site 2 showing significantly higher counts ($p < 0.05$). Dominant bacteria included: *Bacillus* spp. *Pseudomonas* spp. *Micrococcus* spp. *Streptomyces* spp.

Spore-forming *Bacillus* species were abundant, reflecting their resilience to environmental stress, mechanical disturbances, and desiccation typical of quarry environments.

Fungi

Fungal counts ranged from 1.7×10^3 to 4.5×10^4 CFU/g, with predominant genera: *Aspergillus niger*, *Aspergillus flavus*, *Penicillium* spp. *Fusarium* spp.

The presence of *Aspergillus* aligns with prior studies reporting airborne fungal spores in open, dusty habitats.

Molecular Identification and Phylogenetic Analysis

Sequencing confirmed the identity of dominant isolates with 97%–100% similarity to known species.

Bacillus subtilis, *Bacillus cereus* and *Bacillus megaterium* formed a major cluster. *Pseudomonas aeruginosa* and *Pseudomonas putida* grouped with environmental strains known for hydrocarbon degradation. Fungal ITS sequencing identified *Aspergillus niger*, *Penicillium chrysogenum*, and *Fusarium solani*. Phylogenetic trees showed genetic relatedness to soil and sediment-derived strains previously reported from disturbed mineral environments.

Table 2: Total Microbial Load Of Quarry Sand Samples

Microbial Group	Site A (CFU/g)	Site B (CFU/g)	Site C (CFU/g)	Media Used
Total Bacteria	1.8×10^6	2.1×10^6	1.5×10^6	Nutrient Agar
Fungi	3.1×10^4	2.8×10^4	4.2×10^4	PDA
Actinomycetes	6.6×10^5	5.9×10^5	7.2×10^5	Starch Casein Agar
Coliforms	1.2×10^3	8.4×10^2	2.3×10^3	MacConkey Agar
Yeasts	4.5×10^3	4.0×10^3	5.2×10^3	SDA

Correlation with Environmental Factors

Bacterial abundance had a **strong positive correlation with moisture** ($r = 0.82$). Fungal abundance showed a **positive relationship with organic matter** ($r = 0.71$). Grain size negatively correlated with microbial load: finer sand retained more microbes due to higher surface area.

These findings demonstrate that microhabitats in quarry sands are shaped by moisture variation, air exposure, and substrate texture.

Table 3: Morphological And Biochemical Characteristics Of Bacterial Isolates

Isolate Code	Shape & Gram Reaction	Catalase	Oxidase	Motility	Presumptive Identity
B1	Gram + Rod	+	–	+	<i>Bacillus</i> spp.
B2	Gram – Rod	+	+	+	<i>Pseudomonas</i> spp.
B3	Gram + Cocci	–	–	–	<i>Staphylococcus</i> spp.
B4	Gram – Rod	+	–	+	<i>Enterobacter</i> spp.
B5	Gram + Filamentous	+	–	–	<i>Actinomyces</i> spp.

Discussion Summary

The quarry sand microbiome was dominated by stress-tolerant bacteria and filamentous fungi. Microbial diversity remained lower compared to topsoil but higher than expected for a low-organic substrate. Key interpretations: Spore-formers (*Bacillus*) thrive due to physical disturbances in quarry environments. Presence of *Pseudomonas* indicates environmental adaptability and potential for biodegradation. Fungal genera (*Aspergillus*, *Penicillium*) suggest airborne deposition. Microbial profiles varied significantly by site due to differences in moisture, sunlight exposure, and quarry activity.

Table 4: Fungal Isolates Observed In Quarry Sand

Isolate Code	Colony Color	Hyphal Type	Spore Type	Presumptive Identity
F1	Green	Septate	Conidia	<i>Aspergillus niger</i>
F2	White	Septate	Conidia	<i>Penicillium spp.</i>
F3	Black	Non-septate	Sporangiospore	<i>Rhizopus spp.</i>
F4	Brown	Septate	Conidia	<i>Cladosporium spp.</i>

CONCLUSION

This study revealed that quarry sand, despite being considered an inert material, hosts diverse bacterial and fungal communities. *Bacillus*, *Pseudomonas*, *Aspergillus*, and *Penicillium* were among the dominant genera isolated from the samples. Physicochemical characteristics such as moisture, pH, and organic matter significantly influenced microbial abundance. Molecular identification confirmed the presence of ecologically and industrially important species with potential roles in biodegradation and soil formation. The findings highlight the need to consider microbial quality when using quarry sand in construction or environmental applications.

FUTURE WORK

To expand this research, the following directions are recommended:

High-Throughput Sequencing

Use next-generation sequencing (NGS) and shotgun metagenomics to obtain comprehensive microbial profiles, including unculturable taxa.

Functional Characterization

Assess enzyme activity, pollutant degradation capability, biosurfactant production, and antimicrobial properties of isolates.

Longitudinal Studies

Monitor microbial succession during quarry sand storage, transport, and use in construction or landscaping.

Comparative Studies

Compare quarry sand microbiomes with river sand, manufactured sand (M-sand), and soil to evaluate ecological differences.

Risk Assessment

Evaluate potential pathogenicity and allergenicity of isolates, particularly concerning workers exposed to quarry dust.

Environmental Application Trials

Test beneficial isolates for use in bioremediation, compost enhancement, and soil restoration in degraded quarry lands.

REFERENCES

- Ahemad, M. (2014). Microbial ecology of soil: A review. *Applied Ecology and Environmental Research*, 12(1), 261–276.
- Basak, B., & Biswas, D. R. (2009). Influence of microbial inoculants on soil biological properties. *Soil Biology and Biochemistry*, 41(6), 1275–1283.
- Beattie, G. A., & Lindow, S. E. (1995). The secret life of the phyllosphere: Microbial diversity and interactions. *Annual Review of Phytopathology*, 33, 145–172.
- Berendsen, R. L., Pieterse, C. M., & Bakker, P. A. (2012). The rhizosphere microbiome and plant health. *Trends in Plant Science*, 17(8), 478–486.
- Bhaduri, A. M., & Deming, J. W. (2013). Microbial diversity in cold mineral substrates. *Frontiers in Microbiology*, 4, 403.
- Caruso, T., Rillig, M. C., & Garlaschelli, D. (2012). On the structure and properties of ecological networks. *PLoS Biology*, 10(8), e1001423.
- Chakraborty, P., Kar, R. N., & Das, S. (2015). Bacterial communities in laterite and mining-affected soils. *Environmental Monitoring and Assessment*, 187, 4175.
- A Muspira, W. Anitha, Swathi T, Jenifer E, L.Ashwini , (2025) Development And Quality Evaluation Of Honey-Flavoured Yogurt Supplemented With Papaya And Grape Pulp, *The Bioscan*, 2020(3): S.I (3), 996-1000
- Revathi K , Harishkumar B , R Lavanya , Linisha.N.M, Maram Soumya Sree , (2025) Honey-Flavoured Probiotic Yogurt Enriched With Fruit Pulp: A Review On Nutritional, Functional And Sensory Perspectives, *The Bioscan*, 2020(3): S.I (3), 992-995
- Senthil Kumar.K.S, Senthilkumar G P , R Lavanya , Linisha.N.M, M. Sudha ,(2025) Emergence Of Green Fungus (*Aspergillus*) In Covid-19 Recovered Patients: Clinical Implications And Preventive Strategies, *The Bioscan*, 2020(3): S.I (3), 987-991

11. Senthil Kumar. K. S, Senthilkumar G P , R Lavanya , Linisha.N.M, Paranthaman , (2025) Selective Cytotoxic Effect Of Allium Ascalonicum Ethanol Extract Against Hepg-2 Cells Via Ros-Mediated Apoptosis, 2020(3): S.I (3), 980-986
12. Steniffer Jebaruby Stanly , Sent Hilkumar G P B Devasena³, Linisha.N.M, Paranthaman , (2025) Activated Carbon-Based Filtrat Io, The Bioscan N Systems: Advances And Applications In Water Purification, The Bioscan, 2020(3): S.I (3), 976-979