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Conservation of Tufa-Environment Microbiota in Jiuzhaigou National Nature Reserve

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ABSTRACT
The unique tufa landscapes of Jiuzhaigou are increasingly threatened by tourism development and global climate change. This study systematically classifies three representative tufa types in Jiuzhaigou and analyzes the functional mechanisms of prokaryotic and eukaryotic microorganisms in carbonate deposition and dissolution. It highlights unresolved issues such as unclear pathways and data insufficiency in microbial-mediated processes. The paper further calls for the integration of molecular mechanism research with ecological management to enhance landscape protection and sustainable development.

KEYWORDS
World Heritage Site; Environmentally Driven Microbial Processes; Calcium Carbonate Precipitation; Ecological Conservation

1 Introduction

Jiuzhaigou is located in the northern part of the Aba Tibetan and Qiang Autonomous Prefecture of Sichuan Province. As a national nature reserve and UNESCO World Heritage Site, it is renowned for its distinctive landscapes comprising tufa-dammed lakes and cascading waterfalls. Since the official opening in 1984, annual tourist visits have surged from 28,000 to 5.85 million by 2024. Ticket sales alone now exceed 640 million yuan annually, and the tourism industry employs more than 80,000 people.

The reserve spans 720 km^2 and ranges in elevation from 1,996 to 4,880 meters. Of the 27 core heritage sites, 12 are characterized by tufa landscapes, where many lake dams are composed of carbonate deposits. Tufa is a typical form of freshwater carbonate sediment whose formation is strongly microbially mediated [1][4]. However, Jiuzhaigou's delicate tufa features are under growing pressure from tourism development and the broader impacts of global climate change.

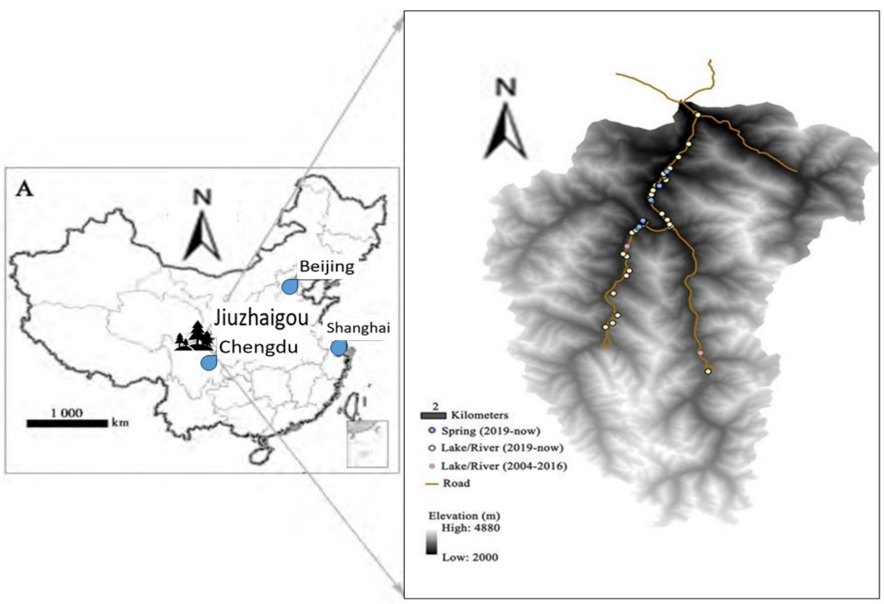


Figure 1

Figure 2 Topographic map and representative view of tufa formations in Jiuzhaigou

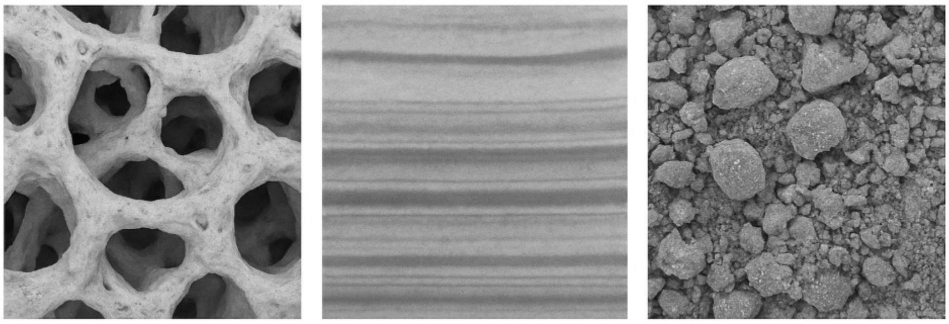
2 Classification of Tufa

Tufa is a freshwater calcium carbonate deposit formed through combined biotic (e.g., cyanobacteria, algae, mosses) and abiotic (e.g., hydrodynamics, hydrochemistry) processes, constructing a characteristic "waterfall–lake" landscape system in Jiuzhaigou [71]. The local tufa accretes slowly—at a rate of mere millimeters per year—and its formation is influenced by hydrology, microbial activity, and geomorphic evolution, often requiring millennia [2][3].

Jiuzhaigou's tufa is typically gray or yellow, while in Huanglong it is predominantly white or yellow. Whiter hues generally indicate less biological involvement. The 12 core tufa sites in Jiuzhaigou exhibit gray-yellow coloration, reflecting biogenic origins. In zones with active microbial processes, higher total nitrogen and phosphorus concentrations and lower calcium oxide levels are observed, resulting in denser, harder deposits [72].

Based on structure and genesis, Jiuzhaigou's tufa is categorized into three types: porous, laminated, and clastic [5]. The primary minerals are calcite (typically abiogenic) and aragonite (often microbial in origin) [12]. These deposits precipitate from $CaCO_3$-supersaturated surface waters following the reaction:

$Ca(HCO_3)_2 \rightleftharpoons CaCO_3\downarrow + CO_2\uparrow + H_2O$ [2].



a. Porous travertine b. Laminated travertine c. Clastic travertine

Figure 3 Representative Types of Travertine Structures in Jiuzhaigou

a. Porous travertine: characterized by loosely cemented voids filled with twigs, algae, and moss; b. Laminated travertine: showing rhythmic microbial banding influenced by seasonal or diurnal changes; c. Clastic travertine: formed in wetland backwaters through the accumulation of detrital $CaCO_3$ mixed with organic debris and sediment.

2.1 Porous Tufa

Widely distributed across Jiuzhaigou and still actively forming, porous tufa is evident at sites like Pearl Shoal and Five-Flower Lake. It features large voids, loose structure, and abundant organic residues—typical of cold-water, biologically mediated deposition [5]. Compared with Huanglong, Jiuzhaigou's porous tufa shows stronger biogenic characteristics [15][16]. Its porosity is categorized into five types: moldic, framework, windowed, boring, and shelled structures [17].

2.2 Laminated Tufa

Laminated tufa has been primarily found at Zangmalongli Valley, Jialijiage Spring, and Jianyan, though it is no longer actively depositing. It accumulates more rapidly than other types and is often used for paleoclimatic reconstruction [5]–[9]. Microbial rhythms control the lamination, which reflects daily, seasonal, and annual cycles. Studies in Europe and East Asia indicate that diurnal changes can produce alternating aragonite–calcite bands, and in Japan, cyanobacterial buoyancy fluctuations generate fine daily laminations [10]–[14].

2.3 Detrital Tufa

Clastic tufa is primarily found at the lake bottoms and distal ends of Jiuzhaigou's major lakes, forming wetlands and marshes through the mixing of detrital $CaCO_3$ with soil and organic matter. These zones provide a water-purifying buffer and show a clear tendency to advance toward the lake center—especially in sites like Arrow Bamboo Lake, Mirror Lake, and Rhino Lake.

The clastic layers often contain high organic carbon content and remnants of algal communities. Freytet et al. [18] classified ten types of aragonite crystal morphologies closely associated with algae, as summarized in Table 1.

Table 1 **Calcite crystal types associated with algal structures**(*Adapted from Freytet & Verrecchia, 1998*[18])

No.	Crystal Morphology	Representative Algae
①	Coarse crystals penetrated by aligned filaments	Nostoc, Calothrix
②	Spherulitic calcite on gelatinous sheaths	—
③	Micritic tube structures from decomposed algae	Dichothrix, Phormidium
④	Coarse crystals intersected by mucilaginous stalks	Hydrurus
⑤	Small plates on algal cell walls	Cosmarium, Pediastrum
⑥	Fan-shaped clusters	Phormidium
⑦	Dendritic and tool-box-shaped crystals on mucilaginous sheaths	Scytonema
⑧	Crystals embedded in soft mucilage and gelatinous stalks	Diatoms, Chlorochytrium
⑨	Lamellar or equant crystals	Tribonema, Scytonema
⑩	Rhombohedral crystals in chains or clusters	Scytonema, Asterococcus, Nostochopsis

3 Environmental Microorganisms Involved in Tufa Deposition

Microorganisms play a pivotal role in tufa formation, including prokaryotes (especially cyanobacteria), eukaryotic algae, and fungi. Their presence and metabolic activities strongly influence carbonate deposition, crystal morphology, and sediment microstructure.

3.1 Prokaryotes

Among prokaryotes, cyanobacteria are the most significant contributors to tufa precipitation. These microorganisms can survive in extreme environments—such as high CO_2 concentrations, intense sunlight, or sulfide-rich waters—and promote $CaCO_3$ deposition through photosynthesis and metabolic alkalinity [19].

Common genera in Jiuzhaigou include Schizothrix, Scytonema, Oscillatoria, Gloeocapsa, and Microcystis [20][21]. These cyanobacteria possess mucilaginous sheaths that facilitate mineral crust formation. The filamentous genus Schizothrix, for instance, is widespread in the Northern Hemisphere and often forms blue-green microbial mats [22][23].

In addition to cyanobacteria, filamentous green algae such as Spirogyra, Anabaena, and Fischerella may contribute to carbonate binding. Over time, their surface calcification alters the visual appearance of tufa formations, often darkening originally white deposits and impacting their aesthetic and touristic value [24]–[26].

3.2 Eukaryotic Algae

Eukaryotic algae inhabit primarily cold-water tufa environments and include diatoms, green algae, golden algae, red algae, and yellow-green algae [27]–[30]. Among them, species from Chlorophyceae (e. g., Chara, Cladophora) and Zygnemataceae (e.g., Desmidium) are particularly common in Jiuzhaigou [27][28].

Diatoms are the most diverse and abundant group, with representatives like Gomphonema and Navicula often colonizing mosses and macrophytes [28]. In Huanglong's Five-Color Pond, Synedra is a major pigment-contributing species [29]. Hydrurus and Aphanothece are occasionally observed in shaded streams and humid microhabitats.

Between 2002 and 2004, 47 algal species were identified in Jiuzhaigou waters, 39 of which were diatoms [31]. In the Huanglong tufa system, diatoms also dominate algal biomass [32]. Wang Quanxi et al. [85] documented 260 algal species in the region, summarized in Table 2.

3.3 Fungi

Fungal involvement in tufa deposition has received relatively little attention. However, studies from both Germany and Huanglong have detected ascomycetes and filamentous fungi—such as Saprolegnia and Stysanus—acting as nucleation sites for $CaCO_3$ crystallization [33][34]. Their potential role in the biomineralization process merits further exploration.

Table 2 **Taxonomic Summary of Common Algae in Jiuzhaigou**(*Adapted from Wang Quanxi et al., 2017* [85])

Algal Division	Number of Genera	Number of Species	Varieties	Forms
Bacillariophyta (Diatoms)	54	160	16	0
Chlorophyta (Green Algae)	24	36	3	1
Cyanophyta (Blue-Green Algae)	16	37	1	0
Euglenophyta	2	11	1	0
Chrysophyta (Golden Algae)	4	5	0	0
Xanthophyta (Yellow-Green Algae)	3	4	0	0
Dinophyta (Dinoflagellates)	3	3	0	0
Cryptophyta	2	2	0	0
Charophyta (Stoneworts)	1	2	1	0

4 Microbial Roles in Tufa Deposition, Dissolution, and Inhibition

Tufa deposition commonly occurs when CO_2-saturated surface water contacts air, leading to degassing, increased pH, and subsequent $CaCO_3$ precipitation. Microbial communities serve as catalysts and regulators in this geochemical process, forming a dynamic feedback loop of precipitation–dissolution (Figure 4).

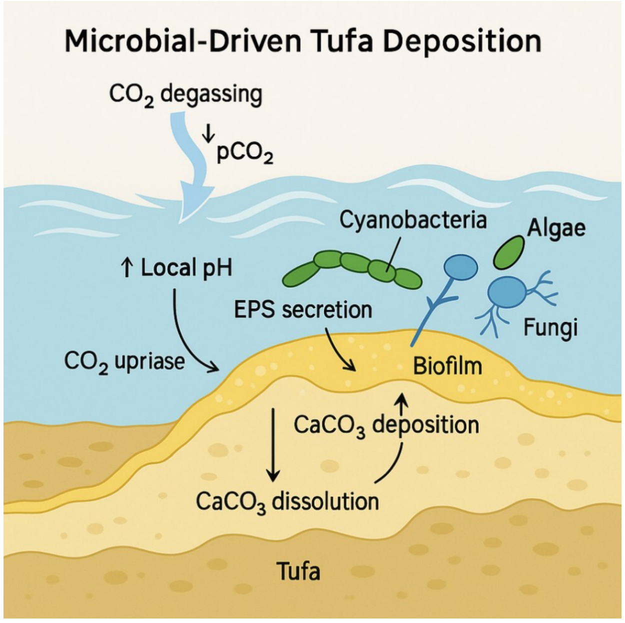


Figure 4 Microbial-Driven Tufa Deposition

4.1 Microbial Contributions to Tufa Precipitation

Since the 16th century, numerous experiments have confirmed that purely physicochemical mechanisms cannot fully explain $CaCO_3$ precipitation; microbial biofilms significantly enhance this process [44][45]. Through metabolic activities—such as photosynthesis, ammonification, denitrification, and sulfate reduction—algae, cyanobacteria, and heterotrophic bacteria elevate local pH and consume CO_2, promoting the formation of aragonite or calcite crystals [46]–[49][51]–[53].

Extracellular polymeric substances (EPS), secreted by these microbes, provide nucleation sites and influence crystal morphology and spatial distribution [54]–[56]. Studies in Huanglong and Yellowstone National Park demonstrate that cyanobacteria are primary drivers of carbonate frameworks, with mineral precipitation occurring around the outer sheath, leaving the lumen hollow and enhancing porosity [50][71].

4.2 Microbial Contributions to Tufa Dissolution and Inhibition

Certain fungi and bacteria can also dissolve $CaCO_3$ by secreting organic acids and enzymes, notably carbonic anhydrase (CA), which catalyzes the interconversion between CO_2 and HCO_3. This enzyme is widely found in microorganisms such as Neisseria, diatoms, and filamentous fungi, and plays a critical role in the bioweathering of carbonate rocks [57]–[61].

Additionally, some algae produce neutral extracellular polysaccharides or organic acids that inhibit crystal growth or alter crystal morphology [62]. For example, Nostoc species, despite abundant EPS secretion, exhibit weak calcification potential due to lack of acidic functional groups [70]. In contrast, green algae can chelate calcium ions through polysaccharides, decelerating precipitation while facilitating aragonite formation or inducing crystal corrosion [72].

4.3 Dual Role of Microorganisms in Precipitation and Dissolution

Microbial communities simultaneously promote and inhibit tufa formation, depending on their biochemical pathways and environmental conditions [65]–[67]. Variability in extracellular enzyme activity and species-specific preferences for mineral phases contribute to the complexity of carbonate dynamics. The composition of microbial mats determines crystal habit, mineral stability, and overall sediment texture [68]–[70].

For example, Schizothrix and Microcoleus commonly precipitate $CaCO_3$ along mucilaginous sheaths, while other cyanobacteria may exert negligible influence, suggesting the presence of biomineralization inhibitors within certain taxa [70].

5 Conclusions

Although numerous studies have explored the microbial mechanisms underlying tufa deposition and dissolution from various perspectives [73]–[84], much of the current understanding remains conceptual and lacks support from modern quantitative and molecular approaches. Key challenges include taxonomic ambiguities, insufficient recognition of "uncultured" microbial taxa, and limited integration of ecological function with geomorphic evolution.

Future research should emphasize molecular biology techniques—such as ancient DNA (aDNA) extraction, metagenomics, and high-throughput sequencing—coupled with machine learning-based modeling to clarify the regulatory pathways and feedback dynamics of microbial-induced $CaCO_3$ formation and breakdown.

To strengthen the conservation of tufa-based ecosystems, we recommend a six-pronged strategy (Figure 5):

Baseline Microbial Surveys and Functional Studies: Establish functional microbial population databases through metagenomic profiling and in situ experiments.

Zonal Protection and Ecological Redline Designation: Define core microecological protection zones and restrict anthropogenic disturbances.

Control of Nutrient Loading in Water Bodies:Monitor total nitrogen (TN) and total phosphorus (TP) concentrations and manage inflow water quality.

Intelligent Monitoring and Early-Warning Systems: Integrate remote sensing, IoT-based sensors, and AI analysis for dynamic ecosystem tracking.

Public Science Communication and Ecological Education: Guide tourist behavior through targeted education campaigns and eco-literacy programs.

Policy Support and Funding Mechanisms: Promote collaboration between scientific institutions, local management agencies, and community stakeholders.

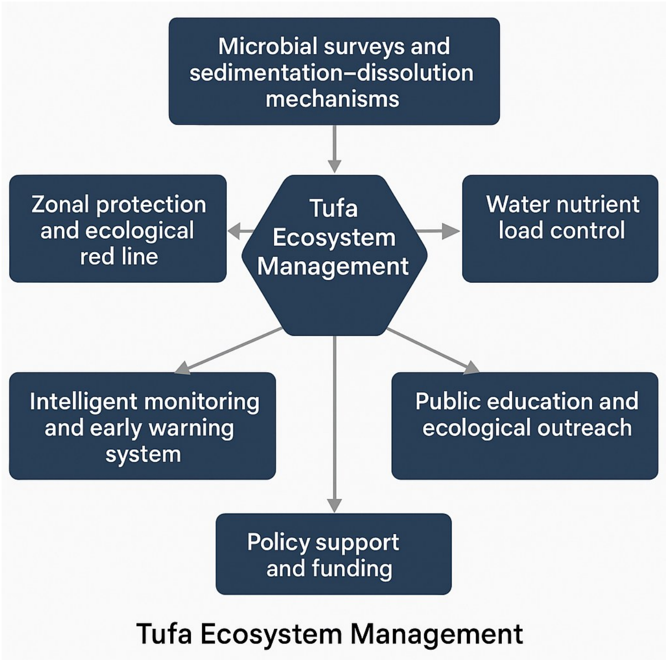


Figure 5 Integrated Strategy for Microbial Conservation in Tufa Ecosystems

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