

A Survey of the Microbial Diversity of an Alkaline Biofilm Associated with a Unique Geological Feature within a Hot Spring of the Hypersaline Alkaline Basin of Lake Magadi, Kenya Jackie D. Denson¹, D. Mack Ivey¹, Stefan Leuko², Lynn J. Rothschild² and Kevin L. Hand³, ¹University of Arkansas, Fayetteville, AR ²Ames Research Center, Moffet Field, CA, ³Jet Propulsion Laboratory, Pasadena, CA. (jdenson@uark.edu),

Introduction: Lake Magadi, Kenya, is an alkaline, "saline pan" lake system, and approximately 100 sq km in size being geographically the southern-most lake within the Rift Valley of Kenya (Fig. 1). Located approximately 80 miles southwest of Nairobi, the lake contains concentrated brines of sodium carbonate as well as large precipitates of the mineral trona, hydrated sodium bicarbonate carbonate ($\text{Na}_3\text{HCO}_3\text{CO}_3 \cdot 2\text{H}_2\text{O}$). The lake water is primarily recharged by meteoric input as well as by a number of saline hot springs (up to 83°C) flowing into the lake's extremely alkaline system (pH values up to ~ 11). This creates a unique extreme environment, containing both thermal, highly saline, and extreme alkaline features all located in close proximity and overlapping to various degrees [1]. While some microbial diversity of this region has been characterized [2], many of the unique springs of the region have yet to be examined.

Here we report the microbial diversity analysis from an alkaline hot spring biofilm associated with a unique erosional texture in the substrata beneath. Amplicons were obtained utilizing universal bacterial (27F and 1525R, 342F and 1407R), archaeal (8F and 1041R), and nanoarchaeal (9F and 961R) universal primers, which were subsequently cloned and sequenced.



Figure 1. Lake Magadi, Kenya

Hot Spring Biofilm: ($2^\circ 00' 24.46'' \text{S}$, $36^\circ 14' 05.80'' \text{E}$) The hot spring complexes of the Magadi region contain a wide variety of niches based on initial observations, including phototrophic mats as well as filamentous structures. One complex located at southern tip of the lake contained a unique structural feature associated with a thin ($>1\text{cm}$ on average) biofilm (Fig. 2). The salinity at this site was $\sim 10 (\text{S} \cdot \text{m}^{-1})$, about twice that of seawater, with a pH of 9.6, and a maximum recorded temp of 45°C .



Figure 2. Biofilm located in alkaline hot spring with terraced erosional structures.

This biofilm was extensively co-located with a "terraced" erosional structure occurring within the surface of the evaporitic deposit from which the spring emerged, suggesting that it is involved in the formation of these erosional structures. These features occurred both in the surrounding margins of the spring as well as within the spring as terraced mound-like features, with the spacing of the terraces appearing to correlate with the slope of the surface to which the biofilm is attached. The springs at this site are known to be nomadic, with locations of flow migrating with time. This could possibly lead to preservation of these structures as biosignatures in the geological record. Chemical analysis of the water from this site as well as community analysis of samples of this biofilm were undertaken in order to gain insight into the formation of these unique features. The chemical analysis of the water from the site is depicted in Table 1, the significant values for metal are listed and were obtained by ICP-OES.

Conductivity (field measurement)	~10 (S•m ⁻¹),
Average pH (field measurement)	9.6
Boron	7.24 mg/l
Calcium	1.00 mg/l
Chloride	6034.10 mg/l
Fluoride	111.59 mg/l
Iron	0.19 mg/l
Lead	0.25 mg/l
Molybdenum	0.11 mg/l
Ortho phosphate	4.62 mg/l
Potassium	174.99 mg/l
Selenium	0.87 mg/l
Sodium	11540 mg/l
Sulfate	214.62 mg/l
Zinc	0.31 mg/l

Table 1. Chemical Analysis of the Spring Water (only values obtained > .20 mg/l are listed)

Diversity Data: Initial analysis of prokaryotic sequences obtained reveals that this biofilm is composed predominantly of a variety of bacteria as well as a number of OTUs (operational transcribed units) related to the marine *Crenarchaeota*. However, among the bacterial OTUs obtained, two groups appear to dominate this system - a number of related sequences of the alphaproteobacteria, most closely related to the photosynthetic genus *Rhodobacter*, and a single OTU most closely related to a *Fusibacter* sp. of the *Firmicutes* known for thiosulfate reduction (Fig. 3). *Rhodobacter* sp. are known to possess an extensive range of metabolic capabilities, including photosynthesis, fermentation, aerobic respiration, lithotrophy to obtain reducing equivalents as well as nitrogen fixation [3]. Given the relative abundance of sulfate in this system, it can be envisioned that the sulfate-reducing *Fusibacter* sp. in combination with the photosynthetic alphaproteobacteria (utilizing sulfides produced through sulfate reduction) are potentially responsible for producing these erosional terraced features [4]. These types of structures could serve as unique geological biosignatures, particularly in sulfate rich evaporitic environments. In addition a variety of other sequences including those of alkaliphilic *Bacillus* and *Clostridia*, as well as some cyanobacterial sp. were found in smaller numbers.

Ongoing Research: Further characterization of the microbial diversity present within this site is currently underway, including the eukaryotic components.

Analysis of the evaporite material has begun, including isotopic studies of the C, O, and S, components of the substrata and biofilm components. This will potentially give us further insight into the formation of these unique features. Future visits to this site will include attempts to locate remnants of dead spring sites in the area in order to see if these features are being preserved geologically.

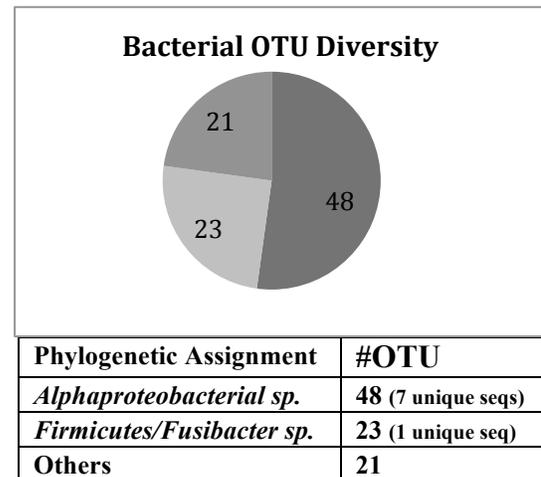


Figure 4. Phylogenetic Assignments of the Bacterial OTUs Sequenced to Date.

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