

INVESTIGATOR'S ANNUAL REPORT

National Park Service

All or some of the information provided may be available to the public

Reporting Year: 2004	Park: Yellowstone NP
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Permit#: YELL-2004-SCI-2160	
Park-assigned Study Id. #: YELL-02160	
Project Title: Molecular ecology of photosynthetic hot spring bacteria that resemble <i>Heliothrix oregonensis</i>	
Permit Start Date: Jan 17, 2004	Permit Expiration Date Dec 31, 2004
Study Start Date: Jan 17, 2004	Study End Date Dec 31, 2004
Study Status: Continuing	
Activity Type: Research	
Subject/Discipline: Microbes	
Objectives: <p>As part of our National Science Foundation Microbial Observatory project, we are currently facilitating 3 projects that involve permitted fieldwork in Yellowstone:</p> <p>(1) Site Monitoring: having characterized several genetically distinct "Red Layer" communities throughout the park, we just completed year 2 of a 5-year monitoring/database project to assess physical and chemical site features (temperature, pH, water chemistry) alongside biological information about 4 red layer communities (pigment analysis, microscopic assessment, DNA/16S cloning/sequencing and DGGE population analysis). We are attempting to address physical/chemical site features that may account for observed site-specific genetic variation.</p> <p>(2) Mat Formation In Situ: having observed Red Layer filaments (Red Chloroflexi) in geothermal groundwater, we have continued studies that attempt to grow and characterize Red Layer mats in situ using sterile glass rods suspended from local tree branches in spring run-off. After one preliminary, small-scale experiment established this could be done at Fairy Spring (2003-4), we set up a more formal experiment with more replicates in June 2004. Although we have retrieved 75% of these rods, our final collection timepoint will be June 2005. In this experiment, we are attempting to address how layered mat formation occurs, assessing which microbes initially colonize, when each group (Cyanobacteria, Green Chloroflexi, Red Chloroflexi, chemotrophic bacteria) establishes detectable colonization, and when distinct layering takes place.</p> <p>(3) New Red Layer Discovery: although we have surveyed many front- and back-country regions of Yellowstone for Red Layer communities, we aim</p>	

to explore a few key remaining additional regions in Yellowstone.

(4) Red Layer Culture Design: Undergraduate Terry Manning, initially supported by an American Society for Microbiology student fellowship in 2003/4, is completing an Honors Thesis to develop and assess site-specific Red Chloroflexi media based on water chemistry data from Hillside Springs.

Findings and Status:

In terms of our specific objectives, we have achieved the following in 2004-5:

(1) Undergraduate teams and I performed 2 assessments in 2004: one in June and one in September. Monitored sites included Hillside Springs, Fairy Springs, Spray Geyser, and Imperial Geyser. These data continue to be archived on our on-line "Red Layer Microbial Observatory (RLMO) Database" (www.wou.edu/rmodb). As has been reported for other sites, we are learning that water chemistry and microbe concentration fluctuates over the course of the summer (one of the reasons we added a second timepoint in 2004). As has been observed in the past by us and other scientists, we observed distinct chemical profiles at different Red Layer sites, lending support our hypothesis that site-specific chemical signatures select for site-specific Red Chloroflexi variants. In 2004, we dramatically improved our population genetic methods (DGGE), developing and implementing a useful battery of controls that can be used to better identify key groups of microbes (representative groups of phototrophs and chemotrophs). Although these were selected based on Red Layer-specific data we have collected since 2000, we hope these controls will be useful for other Yellowstone researchers given that many selected controls appear to be ubiquitous components of other alkaline mat systems. As with other monitored data, DGGE profiles are available for view on our RLMO Database (unlike other monitored data, DGGE data was only introduced in 2004).

(2) Mat Formation In Situ: Undergraduate Jennifer Esparza began to analyze 2004 rod replicates in September 2004 and will complete an Honors Thesis about her work (due in 2006). To date, she has analyzed 2 rod timepoints (6 vs. 8 weeks post set-up) in terms of the following: quantitatively characterizing biofilm accumulation, microscopically counting two key groups of phototrophs (Chloroflexi filaments and Cyanobacteria - both using fluorescent techniques), enumerating chemotrophic bacteria using 4 common media/plating approaches, and characterizing extracted pigments. DNA-based methods are in progress at this time. Preliminary data suggests that visible green phototrophs establish the first visible biofilm, along with a diverse population of chemotrophic microbes. Although Chloroflexi filaments are present at moderate levels, they are visible only microscopically - consistent with Jenn's working hypothesis that a visible Red Layer cannot form until a green layer "shield" has established, reducing light intensity and oxygen levels.

(3) New Red Layer Discovery: Over 2004-2005, we performed no additional surveys.

(4) Red Layer Culture Design: Although Terry's project yielded many characterized chemotrophic bacteria and one Green Chloroflexi (a relative of the Red Layer filament), Terry was never able to retrieve Red Layer filaments in culture (to date, the only cultured relative remains Roseiflexus, from Japanese hot springs). All of Terry's retrieved sequences are available on the RLMO database; his Honors Thesis will be complete at the end of 2005.

For this study, were one or more specimens collected and removed from the park but not destroyed during analyses?

No

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Fill out the following ONLY IF the National Park Service supported this project in this reporting year by providing money to a university or college

Full name of college or university:

n/a

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