Annual Report for Period:02/2004 - 02/2005 **Submitted on:** 12/13/2004

Principal Investigator: Boomer, Sarah M. **Award ID:** 0237167

Organization: Western Oregon University

Title:

RUI-Microbial Observatories: A Longitudinal Molecular Diversity and Chemical Survey of Red Layer Microbial Communities in Yellowstone National Park

Project Participants

Senior Personnel

Name: Boomer, Sarah

Worked for more than 160 Hours: Yes

Contribution to Project:

In addition to continuing to facilitate previously-described research-based undergraduate course lab experiences in General Microbiology and Computational Biology, I have facilitated the following new projects based on specific goals in the renewed grant (to be described in more detail in other sections): (1) developed and implemented new summer undergraduate research program GERMS, Geochemistry and Ecology of Red Mat Systems; (2) developed and implemented 6 new microbial diversity/biotechnology workshops for secondary science teachers; (3) managed and advised the development and implementation of pre-college outreach activities in microbial diversity and biotechnology (Kelly Shipley served as the primary instructor, coordinator, and assessor) for Upward Bound, Saturday Academy, and the new WAMS program (see next item); (4) Co-Direct new campus-based pre-college outreach program WAMS, Western Adventures in Math and Science (with Kelly Shipley and 3 other Science/Math Faculty); (5) contributed to the authorship of 3 ASM General Meeting posters (one undergraduate research and two curriculum-based); (6) co-authored a book chapter about RLMO Database and one manuscript about Computational Biology curriculum (rejected by ASM Microbial Education 11/04 but revised/submitted to the National Science Teachers Association 12/04); and (7) advised or am advising two undergraduate Honors Research/Thesis projects (Terry Manning and Jennifer Esparza).

Name: Shipley, Kelly

Worked for more than 160 Hours: Yes

Contribution to Project:

After completing her Upward Bound cohort project in March 2004 and her MAT in June 2004, Kelly worked part-time from June to August 2004. During this time, she trained with Danny Lodge (former research assistant, also part-time during this period) and facilitated the first undergraduate GERMS program. Kelly began full-time at the end of August 2004, effectively spear-heading the successful development and advertising of our new campus-based pre-college science and math outreach program, WAMS (see Activities/Findings for more information). In addition to working with WAMS and Upward Bound, she has provided pre-college outreach modules for Saturday Academy and GEAR-UP program (again, please see Activities/Findings). Kelly has been a tremendous asset, not only for RLMO-driven outreach û but also to the entire Division of Natural Science and Math.

Name: Dutton, Bryan

Worked for more than 160 Hours: No

Contribution to Project:

Bryan, as described in the Final Report for our last MO grant, has been an outstanding colleague and collaborator, assisting with curriculum assessment and revision, phylogenetics, and û more importantly û database design. Bryan serves as one of the WAMS Co-Directors and has designed and implemented a regular series of pre-college outreach modules for this program (all gratis). Bryan co-authored 2 ASM General Meeting Posters, the RLMO Database chapter (in press), and the Computational Biology Manuscript (in revision).

Post-doc

Graduate Student

Undergraduate Student

Name: Students, Microbiology

Worked for more than 160 Hours: No

Contribution to Project:

A total of 28 undergraduates participated in the revised Microbiology curriculum in Spring and Fall 2004. As proposed and outlined in this grant and described in last year's annual report, students mastered five weeks of traditional culture-based environmental enrichment and phenotype-based identification testing followed by five weeks of culture-independent molecular methods using the RLMO project. Students used lab-dedicated computers to assemble web pages abut their projects, incorporating all new digital technology in the lab. In terms of new revisions to this curriculum, we have expanded molecular detection/identification methods to include pigment analysis and fluorescence microscopy-based detection methods using RLMO samples.

Name: Drury, Will

Worked for more than 160 Hours: No

Contribution to Project:

Will Drury, supported as a summer research student in 2003, has continued to assist with RLMO database modifications - funded as needed by 'Computer Services' funds. Specific upgrades Will developed for the database this year include new data entry fields that allow us to describe DNA extraction methods and PCR amplification methods for RLMO clones. Correspondingly, Will developed query boxes for searcing the database using these new parameters. Finally, he co-authored a major book chapter about the RLMO Database (in press).

Name: Manning, Terry

Worked for more than 160 Hours: Yes

Contribution to Project:

After receiving one of two RLMO research stipends for June 2002-3, Terry went on to earn one of 25 national ASM summer research fellowship for a continuation of this work (June 2003-4). Terry used site-specific chemical information to develop designer media, comparing retrieval rates using designer media with those using Hanada's Roseiflexus/Red Chloroflexi media (see Activities/Findings). In the past year, Terry completed research toward his Honors Thesis and developed and presented an ASM poster at the General Meeting (May 2004). He submits his Honors Thesis for committee review in January, 2005.

Name: Esparza, Jennifer

Worked for more than 160 Hours: Yes

Contribution to Project:

As a result of participating in GERMS (please see full description of this undergraduate research program in Activities/Findings), Jennifer asked to work with me on her Honors Thesis, specifically on mat formation studies (see Activities/Findings). In September 2004, Jennifer participated in a second field trip to Yellowstone, performed monitoring studies on long-term RLMO sites, collected specific materials for her Thesis work, and attended the Microbial Observatory workshop with Kelly, Terry, and I. Using preliminary data, Jenn developed a research plan that was recently approved by the campus Honors Committee. Jennifer will graduate in June, 2006 and hopes to attend graduate or medical school following graduation.

Name: Brooks, Jana

Worked for more than 160 Hours: No

Contribution to Project:

Jana, a senior/integrated science major when she participated in GERMS (please see full description of this undergraduate research program in Activities/Findings), completed her degree and is now a research assistant at Oregon State University. Her current project position combines both immunology and ecology using a fish model.

Name: Jacobsmuhlen, Elizabeth

Worked for more than 160 Hours: No

Contribution to Project:

Elizabeth, a senior/biology and criminal justice major when she participated in GERMS (please see full description of this undergraduate research program in Activities/Findings), completed her degrees and currently works for the Department of Immigration. She is considering applying for medical school.

Name: Smith, Monica

Worked for more than 160 Hours: No

Contribution to Project:

Monica, a junior/biology major when she participated in GERMS (please see full description of this undergraduate research program in Activities/Findings), is completing her biology degree at this time. She is completing plant physiology research with

colleague Dr. Lonnie Guralnick and intends to pursue a doctoral degree after graduating.

Name: Quezada, Maria

Worked for more than 160 Hours: No

Contribution to Project:

Maria, a freshman/biology major when she participated in GERMS (please see full description of this undergraduate research program in Activities/Findings), is completing her biology degree at this time. She intends to apply to medical school when she graduates.

Name: Howell, Nathan

Worked for more than 160 Hours: No

Contribution to Project:

Nathan, a sophomore/biology major when he participated in GERMS (please see full description of this undergraduate research program in Activities/Findings), is completing his biology degree at this time. He intends to apply to medical school when he graduates.

Name: Students, Computational

Worked for more than 160 Hours: No

Contribution to Project:

A total of 8 biology majors were involved with 5 weeks/30 hours of bioinformatics-based computational biology methods (16S rRNA analysis and phylogenetics plus protein structure determination and modeling). Given that we were developing a manuscript about this work (which included assessment that would be compared to previous course offerings), we made no major curriculum revisions. Although the poster-based presentation of this work was well-received (ASM General Meeting, May 2004 - available on-line at our RLMO website), the submitted manuscript to Microbial Education in October 2004 (also available on-line) was rejected on the basis that it was too methods-heavy and too impact/assessment-light. More information about this curriculum, manuscript, and future plans can be found in Activities/Findings.

Technician, Programmer

Name: Lodge, Danny

Worked for more than 160 Hours: Yes

Contribution to Project:

From February-August 2004, Danny worked as a part-time technician; I retained his previous job description (70% research and 30% teaching) during this time. From February-May, Danny developed new DGGE controls (see Activities/Findings), performed preliminary assessments of year one mat formation studies (see Activities/Findings), and assisted in the teaching of Computational Biology (Bi301). For the latter, Danny was the first author and presenter of a well-received curriculum poster at the ASM General Meeting (May 2004); he also co-authored a manuscript about this course (currently being revised). From June-August, Danny worked with Kelly and I to facilitate the first undergraduate GERMS program and trained Kelly in general research and ordering tasks. In September, Danny began his advanced degree in engineering at OSU. We remain in touch with Danny for manuscript development and technical advice.

Name: Kernan, William

Worked for more than 160 Hours: No

Contribution to Project:

Bill, as described in last year's report, has been integral in the maintenance and support of the RLMO Database. Bill continues to serve as Will Drury's direct supervisor. As Director of Campus Computing Services, Bill will continue to oversee computer service projects as carried out by students like Will (who receive the only direct support) as this aspect of the grant continues.

Other Participant

Research Experience for Undergraduates

Organizational Partners

Upward Bound, Western Oregon University

Upward Bound is a college preparatory program designed to supplement high school programs with college experience in a variety of classes as well as provide counseling, tutoring and a positive environment for students. The program is designed for minority, low-income students and first generation college attendees. Our partnership was facilitated through Western Oregon University's Upward Bound program (Sandra Hastings, Director). This year, 6 pre-college Upward Bound students completed a monthly outreach program with Kelly Shipley, emphasizing microbial diveristy and biotechnology. Kelly assembled web-pages of student work and students were able to access and analyze data on-line from their respective high schools.

American Society For Microbiology

ASM provided Terry Manning (research student June 2002-present) with one of 25 national undergraduate summer research fellowships. Terry received most of this stipend for thesis-related research over summer 2003; in May 2004, he was funded to travel to New Orleans where he presented his research at the ASM General Meeting. He is currently completing his Honors Thesis.

Saturday Academy, Oregon University

Saturday Academy is a national non-profit program that provides fee-based college-level enrichment experiences for pre-college students (grades 7-12). The only local Saturday Academy program is hosted by Oregon State University. Fees, all of which go to OSU/Saturday Academy, facilitate advertising, recruitment, and registration.

Western Adventures in Math and Science

In February 2004, colleagues and I began developing and defining this new non-profit, on-campus program: Western Adventures in Math and Science (WAMS). WAMS aims to be a self-sustaining long-term program, offering low-cost science and math activities and need-based assistance/scholarships to local community members. Since the formal beginning of WAMS activities (March 2004), 14 of 30 Natural Science and Math faculty have enthusiastically devoted time (all gratis) to offering WAMS activities, providing 3-5 hour, mostly weekend workshops in biology, earth science/geology, forensics, and math. In total, 11 WAMS workshops have served 79 students.

GEAR-UP

In November 2004, we began partnering with GEAR-UP (Gaining Early Awareness and Readiness for Undergraduate Programs). GEAR-UP is a partially funded grant program through the Department of Education, serving 19 high schools and 22 middle schools in Oregon, including Monmouth's own Central High School. GEAR-UP's primary goal is to ensure that Oregon's low-income middle and high school students are prepared for, pursue, and succeed in postsecondary education. Kelly Shipley has begun serving approximately 150 freshman students at Central High School in Monmouth, providing activities in DNA isolation and analysis. Future partnership activities include supporting an after-school science club, bringing interested high school students to WOU for first-hand experience in our lab with our RLMO research project, and providing professional development for secondary science teachers so they can adapt microbiology and biotechnology curriculum beyond our presence in the classroom.

Other Collaborators or Contacts

Bev Pierson (U. Puget Sound, Tacoma, WA)- research advice
Tim McDermott (TBI/MSU, Bozeman, MT) research/culturing advice
Bill Inskeep (TBI/MSU, Bozeman, MT) - RCN grant
Mark Young (TBI/MSU, Bozeman, MT)- MO Workshop organization/grant
Maria Fung (WOU Math, Monmouth, OR) - WAMS Co-Director
Phil Wade (WOU Earth Science, Monmouth, OR) - WAMS Co-Director

Activities and Findings

Research and Education Activities:

As described extensively in the next several sections (and elsewhere in this report), this project serves undergraduate research and provides research-driven lab curricula and workshops that provide research-driven experiences for undergraduate biology and/or education majors, secondary science teachers seeking professional development or masters degrees (in education), and pre-college students. Some specific revisions that pertain to last year's annual report:

Geochemical Testing Methodology: Previously acquired geochemical testing equipment has been newly and successfully implemented in both

the new summer GERMS program for undergraduates (including data entry into the RLMO database) and some new workshops for secondary science teachers. We continue to employ equipment collaboratively in majors-level Microbiology (Biology 331) and some pre-college outreach activities.

DGGE Methodology: Following major improvements (described below), previously acquired DGGE equipment has been newly and successfully implemented in the new summer GERMS program for undergraduates (including digital imaging of data on the RLMO database). We are currently developing DGGE-based activities for new teacher workshops. Between February-June 2004, former research assistant Danny Lodge trouble-shot several problems that had been frustrating us with our 2003 experiences using DGGE. Namely, we (mostly Terry Manning, during his research) were spending excessive time and energy cloning/sequencing individual DGGE bands that did not correspond with Roseiflexus or Chloroflexus control bands. Knowing that it would not be physically possible to incorporate such methods into GERMS or future Molecular Biology (Bi475) course offerings, we used a combination of Terry's data and our RLMO clone database to define most-observed non-Green Nonsulfur(GNS)/Chloroflexi phylum members. In addition to GNS/Chloroflexi controls, we now run experimental samples against a new battery of controls that includes RLMO-derived Gram Positives, Proteobacterial subgroups, and Cyanobacteria. During this troubleshooting project, Danny also was interested in assessing variant Roseiflexus-like bands (slightly higher or lower than our Red GNS/Chloroflexi control, Roseiflexus) that we often observed in RLMO-derived experimental samples. Thus, he performed band cloning/sequencing of variants and compared results to our RLMO clone database. To our surprise at the time (it now seems obvious), he was able to show that these DGGE band variants correspond to phylogenetically distinct Red-GNS/Chloroflexi subgroups we had observed and published in 2002. Thus, we have demonstrated that DGGE is an appropriate tool for assessing sub-variation within the Red-GNS/Chloroflexi sub-group.

The Digital Molecular Microbiology Lab: Previously acquired dedicated lab computers, projection systems, sequencing software upgrades, and digital cameras have, in 2004, continued to serve undergraduates in Microbiology and secondary science teachers via RLMO-supported workshops. I continue to be VERY PLEASED with these new systems because they have greatly improved teaching concepts and methods to larger groups of students. A poster presentation of these curricula û emphasizing culture-independent methods and the use of computer/digital technology for lab assignments and presentations û was given by me at the ASM General Meeting in May, 2004 (available on-line at the RLMO website).

Findings:

Major research findings include three areas of progress, all of which have been positively impacted by DGGE and the development of improved RLMO-derived control sequences: (1) the collection of site and community information from four long-term monitoring sites in Yellowstone (the basis for our new undergraduate summer research program, GERMS); (2) the development and assessment of designer media based on site and community information (completed Honors Thesis project of undergraduate Terry Manning); and (3) mat formation studies (newly-initiated Honors Thesis project of undergraduate Jennifer Esparza).

Summer monitoring activities involve several methods and findings we have described previously (site/community characterization, water/mat chemical analysis, water/mat microscopic evaluations, pigment assessment, fluorescence microscopy, and 16S rRNA library analysis from red layer material). Owing to geochemical data presented at the outstanding 'Geothermal Biology and Geochemistry in Yellowstone National Park' meeting (organized by TBI/MSU, October 2003), we expanded our summer monitoring to include 2 sampling timepoints: one at the end of June (all assessments performed by GERMS students) and the second mid-September (fieldwork performed by long-term research students - this year Terry Manning and Jenn Esparza - with molecular studies providing reagents for course-based project during the academic year). We have also expanded analyses to include both the red and green layers, our aim to characterize the extent to which red layer filaments co-inhabit the green layer. Preliminary site and water/community assessments continue to show site-specific chemical profiles. In comparing early vs. late water chemical data, we have noted concentration differences that reflect data presented at the aforementioned meeting and are presumed to be the product of decreasing water run-off as snow melts and typically drier summer conditions prevail. Pigment analysis, microscopy, and DGGE from June/GERMS samples have all demonstrated that red GNS/Chloroflexi are readily present in green layer samples. Interestingly, however, green layer populations contain some members that do not correspond to red layer-derived members (i.e. red layer DGGE control lanes). Unfortunately, given the difficulties with macroscopic dissection and potentially different student handling/perceptions, these data are preliminary. Given GERMS data from the June sampling trip, our more advanced/experienced September team performed more fine layer analysis on the Hillside red layer community, sub-sectioning this red layer into three different zones. Molecular analysis of these samples will provide the basis for Molecular Biology lab this spring.

Terry Manning will submit his Honors Thesis for review in January 2005, describing his 2-year project to develop and test media using site-specific chemical data and spring water from Hillside Springs. Terry received some additional support (both travel and research funding) for this work from the ASM Undergraduate Research Fellowship program. In advising this thesis, I made certain that Terry's project did not

solely rest on the successful cultivation of Red GNS/Chloroflexi because, to date, only one such organism (Roseiflexus, Hanada et al., 2002) has been cultivated despite years of attempts (including by myself as an undergraduate). Over the summer of 2003, Terry used 2 years of collected chemical data (all salts and metals) from Hillside Spring to design 2 different designer media: (1) Hillside-specific chemicals plus high yeast (0.1%) carbon source; and (2) Hillside-specific chemicals plus low yeast (0.001%) carbon source. He compared retrieval rates using these media with those from Hanada's Roseiflexus media (notably 1% yeast extract plus additional chemicals) and filter-sterilized thermal spring water collected at Hillside (recommended by Tim McDermott, TBI/MSU). Using fresh Hillside red layer suspensions, Terry inoculated 8 liquid broth replicates of each media type and incubated in light/50?C under anoxic conditions. Every week for one month, Terry sacrificed 2 replicates for analysis (microscopy, pigment, 16S rRNA cloning, and DGGE). In cases where red/orange growth was visibly occurring/evident, Terry subcultured filaments onto corresponding agar plates. Given the extensive data Terry has gathered (amounting to nearly 500 clones), I will summarize with the following generalizations: (1) Hanada media was consistently and rapidly taken over by consumers (many groups of Proteobacteria) and never yielded promising pigmented enrichments or pure cultures; (2) Both site-specific media selected for consumers, albeit a different subset (most Gram Positives and Acidobacteria); (3) Sterilized spring water consistently retained and supported visible red/orange filaments. Although spring water tubes appeared macroscopically cleaner/clearer than other white/cloudy enrichments, microscopic and molecular assessment revealed a high prevalence of mixed consumers. Spring water streak-plates from pigmented enrichments propagated a co-culture of yellow Chloroflexus-like filaments and Geobacillus-like rods. Encouraged by these results, Terry repeated enrichment studies in 2004, retrieving a pure culture of yellow/orange Chloroflexus-like filaments. In both cases, retrieved Chloroflexus most resembled a unique strain obtained in co-culture by Ward et al. from the 'New Pit' site in Yellowstone. Terry presented these data in May 2004 at the ASM General Meeting in New Orleans.

In July 2003, we began a pilot study to study whether layered photosynthetic mat systems with red layers and/or red layer organisms could be effectively 'grown' in situ using sterile glass rods that were partially submerged in thermal run-off channels of known RLMO sites. It should be noted that Yellowstone was extremely reluctant to approve this study because we asked to leave materials in the field for 1 year. The only study site for which we were approved was Fairy Spring, owing to its distant, off-trail status. We had planned to retrieve subsets of rods in early September and late October but weather and wildlife problems blocked the latter collection date. Danny performed preliminary assessments of the 6-week September set in early 2004, characterizing the early population using DGGE, pigment, and fluorescence microscopy. Although unicellular Cyanobacteria were the most visibly numerous phototroph using pigment and microscopic techniques, an abundance of both green and red GNS/Chloroflexi had already established. These data convinced Yellowstone to let us set up a more careful timecourse series starting in late spring. In May 2004, Danny returned to collect all remaining pilot study rods and re-set-up a new set of replicates. The 9-month replicates had visibly layered communities which contained well-developed green and red layers (as further confirmed via the aforementioned techniques). During the GERMS program, Jennifer Esparza was so taken with the rod experiment when we collected the first set of new replicates that she immediately asked to work on this project for her Honors Thesis. She returned to Yellowstone again in September 2004 to collect the second set of replicates. In November 2004, Jenn's Honors Thesis proposal was approved. Briefly, she is using microscopic methods, pigment analysis, and DGGE to characterize and compare rod populations over time. She has formulated an appropriate hypotheses to test: that Cyanobacteria will be the most abundant phototroph in early rod biofilm communities because GNS/Chloroflexi are known to perform photosynthesis better under low light and oxygen; only after Cyanobacteria have formed a shielding layer will anoxygenic phototrophs accumulate visible layers. I am extremely pleased with Jennifer's initiative, enthusiasm, and rapid grasp of and curiosity regarding potential relationships between metabolism and community structure.

Training and Development:

Course-Based Undergraduate Training: 36 undergraduates were involved in course-based lab curriculum modules using RLMO data and materials. These included the following: (1) 5 weeks/24 in-lab hours of molecular microbiology laboratory methods (16S rRNA library analysis, DNA sequencing, fluorescence microscopy, and introductory computational biology), serving 28 biology majors. Students developed web-based portfolio presentations of this project (available on-line at my RLMO website). These curricula and their assessment provided the basis for a well-received poster at the ASM General Meeting (May 2004, available on-line) and a publication in preparation. (2) 5 weeks/30 in-lab hours of bioinformatics-based computational biology methods (16S rRNA-driven DNA analysis and phylogenetics plus protein structure determination and modeling), serving 8 biology majors. These curricula provided the basis for an extremely well-received ASM poster (May 2004, available on-line); the submitted publication (to Microbial Education in October 2004, with a draft manuscript available on-line), however, was rejected on the basis that it was too methods-heavy and too impact/assessment-light. Dr. Bryan Dutton (who teaches the first 5 weeks of this course) and I are submitting a manuscript proposal of this work to the National Science Teachers Association (NSTA), specifically for a book chapter for their 'Handbook of College Science Teaching: Theory, Research, and Practice.' Dr. Dutton and I are also discussing revamping Computational Biology into the existing Evolution course (which currently lacks a lab component) as a mechanism for increasing student exposure to these important methods.

Independent Research Credit/Thesis Training: 8 undergraduates were involved in independent research credit/thesis training. Of these 6 were admitted to the newly funded summer GERMS program, a 5 week/120 hour research experience that included 1 week in Yellowstone performing site analysis (water chemistry and collection/filtration, site/mat assessment and sample preparation), pigment assessment,

microscopy (light and UV/fluorescence), molecular approaches (16S rRNA clone library assessment and DGGE), and computer-based methods (NCBI/BLAST analysis, database entry, and web-based portfolio development - all projects of which are on-line). Two additional undergraduates, both WOU Honors Students, are also completing senior thesis projects at this time: Terry Manning wrapped up his project to use site chemistry data to develop and test designer media, his hopes to improve culture-based retrieval of Roseiflexus-like Red-GNS. Terry received additional funding from ASM that, this year, included supporting travel to the ASM General Meeting where he authored and delivered his first research poster (available on-line). Terry is completing his Honors Thesis at this time. Jennifer Esparza began working on her Honors Thesis project this summer, having become interested in our ongoing project to study mat formation in situ. As described in the previous section, her Thesis proposal with preliminary data was approved in November 2004. Jennifer will also be submitting an ASM Undergraduate Research Fellowship proposal in February. Both Terry and Jennifer also accompanied me to the NSF-MO Workshop in September 2004.

Graduate/Secondary Educator Training: 21 secondary science educators, all seeking credits toward education masters degree options, have worked with us via formally WOU-credited workshops and activities. In the past year, I have provided 5 one-credit/10 hour weekend workshops covering biotechnology, microbial diversity, and computational biology. Kelly has significantly revised the way we provide teacher workshops by adding both adaptable/low-cost curriculum substitutions that can be used by teachers lacking specific equipment/resources, providing low-cost resource lists to accompany research procedures, and leading discussion groups at each workshops about these topics. Kelly is currently developing a grant resource list to provide and discuss at all workshops as a result of specific feedback from local high school grant-writers and interested science teachers. Because my salary is covered by NSF/MO-provided release time, we are able to provide these workshops at a substantial reduction ($45/credit). In addition, Kelly is also involved in discussions about professional development activities with local teachers involved in the GEAR-UP program and we are exploring non-credit options for professional development training because, despite the low cost of existing credited workshops, it still appears to be detrimental to participation.

Outreach Activities:

With the full-time addition of Kelly Shipley, a Biology and MAT-trained science coordinator (also a former student of mine from WOU), our outreach has expanded significantly. In addition to maintaining ongoing partnerships with Upward Bound and Saturday Academy, Kelly has successfully launched WAMS (to be described below) and begun a major new partnership with GEAR-UP (to be described below).

In terms of previously described partnerships, I want to reiterate that Upward Bound is a national organization that provides free or reduced-cost college-level enrichment experiences to high school students who are minorities, low-income, and/or first-generation college-bound. WOU has a grant-supported Upward Bound program who has facilitated the provision of appropriate cohorts for our work. Kelly completed a series of microbial diversity/biotechnology monthly weekend workshops for 6 Upward Bound students from local high schools. Of these participants, 5 were Hispanic female sophomores and juniors. Kelly's Upward Bound program mirrored the curriculum used in college-level Microbiology (Bi331). Following each activity, Kelly assembled web-pages of student data, which students were able to access and analyze on-line from respective high schools. Kelly also performed and analyzed assessment for this program.

Also in terms of previously described partnerships, Saturday Academy is a national non-profit program that provides fee-based college-level enrichment experiences for pre-college students (grades 7-12). The only local Saturday Academy program is hosted by Oregon State University. In partnership with Saturday Academy, Kelly provided one four-hour class, serving 12 local high school students (1 of whom was a Hispanic male). For this activity, students worked with RLMO-derived clones, performing plasmid isolation and RFLP fingerprinting analysis. There was a notably long waiting list of students interested in the class. Consequently, we will be offering several versions of this class for Saturday Academy in 2005.

Although 'we' (I and many colleagues in the Division of Natural Science and Math - NSM) appreciate partnership opportunities provided by Saturday Academy, we have long been concerned that this fee-based OSU program fails to serve many local students because their typical courses cost $30-$100 (none of which instructors see, for the record). Consequently - as originally proposed - Kelly's position provides 10% effort toward advertising and facilitating outreach connections on behalf of the Division of Natural Sciences and Math - our collective goal to develop a sustainable and sustained on-campus outreach program that would better serve a larger audience of local students. In January 2004, Bryan Dutton and I solicited Division input on this project, with support from appropriate campus administrators. By February, we defined our new non-profit WAMS Program (Western Adventures in Math and Science) and named its Board of Directors: 4 NSM Faculty (Bryan Dutton/Botany and Systemics, Maria Fung/Math and Probability Theory, Phil Wade/Earth Science Education Specialist, and Sarah Boomer/Microbiology and Biotechnology) plus Kelly. Kelly's well-defined role in this program is to solicit interested faculty, collect workshop descriptions using an on-line template we developed, use this information to design brochures, and facilitate advertising (mailing, physically visiting local schools, and submitting information to local school newsletters) - three times a year. With backing from our Dean, staff at the campus Division of Extended Programs manages registration and facilitates printing/mailing. Because we want WAMS to be a self-sustaining long-term program, we debated significantly on an appropriate starting fee and eventually agreed to $20 per activity, our hopes to eventually fully cover advertising costs and to develop a need-based assistance/scholarship program so we could specifically recruit such participants. As proposed, $300 seed money from my grant was initially used to support our first term's advertising costs.

In what has been an overwhelming response, 14 of 30 NSM faculty have enthusiastically devoted time (all gratis) to offering activities for WAMS, providing 3-5 hour mostly weekend workshops in biology, earth science/geology, forensics, and math. Since March 2004, we have run 11 workshops and served 79 students. Although initially conceived as a secondary-level outreach program, we have attained surprising impact numbers with our elementary/middle school-targeted workshops and a higher ratio of such activities are now being offered each term. By September 2004, we were financially in the black and are now accumulating said assistance/scholarship and actively recruiting members of cohorts (like Upward Bound and GEAR-UP) to participate free in this program. Of these WAMS workshops, 4 microbial diversity/biotechnology activities were run by Kelly, serving 15 students. We have also received excellent press in local and statewide newspapers. Of all my grant activities during this transitional year (i.e. replacing a research-oriented technician with a research-oriented outreach position), mentoring Kelly's successful launching of WAMS has been the most productive and successful thing I am proud to have achieved. I and my NSM colleagues sincerely thank NSF for the opportunity to finally develop what we believe will be one of the most significant and long-overdue new programs to our campus and community. As I predicted when I originally wrote this grant, WAMS has truly been a amazing example of 'if you build it, they will come.' Thank you.

As WAMS has gained its own inertia, Kelly has also moved her outreach efforts toward specific partnerships with local high schools, developing RLMO-driven modules for use in the classroom (e.g. for class visits and teacher training/professional development on-site, all facilitated by Kelly). In particular, we began partnering with GEAR-UP (Gaining Early Awareness and Readiness for Undergraduate Programs) in November 2004. GEAR-UP is a partially funded grant program through the Department of Education, serving 19 high schools and 22 middle schools in Oregon, including Monmouth's own Central High School. GEAR-UP's primary goal is to ensure that Oregon's low-income middle and high school students are prepared for, pursue, and succeed in postsecondary education. Kelly has begun serving approximately 150 freshman students at Central High School in Monmouth, providing activities in DNA isolation and analysis. These and future activities are designed for delivery on-site in their classroom, during the normal school day. Future partnership activities include supporting an after-school science club, bringing interested high school students to WOU for first-hand experience in our lab with our RLMO research project, and providing professional development for secondary science teachers so they can adapt microbiology and biotechnology curriculum beyond our presence in the classroom.

Journal Publications

Books or Other One-time Publications

Sarah M. Boomer, Will M. Drury, Bryan E. Dutton, Daniel P. Lodge, Melissa S. Boschee, and William M. Kernan, "The Red Layer Microbial Observatory Database: A Model for the Integration and Dissemination of Biological and Geochemical Data via the World Wide Web", (2004). Book, Accepted
Editor(s): Thermal Biology Institute, Montana State University
Collection: Proceedings from the First Annual Geothermal Biology and Geochemistry in Yellowstone Meeting
Bibliography: Meetings Proceedings Chapter

Terrance F. Manning, "Attempts to Cultivate Roseiflexus-like Bacteria Using Site-Specific Water and Chemistry Data From Hillside Springs, Yellowstone National Park", (2005). Thesis, Submitted
Collection: WOU Honors Thesis
Bibliography: Western Oregon University Honors Department

Web/Internet Site

URL(s):

www.wou.edu/~boomers/research/allresearch.html

Description:

This is our comprehensive RLMO website. It contains RLMO-generated educational course curriculum, outreach projects and assessment, project methodology, and links to our Oracle-based RLMO Database.

Other Specific Products

Product Type:

Data or databases

Product Description:

The RLMO Database Project is a web-accessible Oracle database that integrates physical, chemical, and molecular data from RLMO sites. This application, in its second year of development, was written using PL/SQL, HTML, and Javascript. Each research site in this study is assigned a unique identifier that is linked to the following tables: Geochemical Data (pH, 15 common salts and metals); DNA Sequence Data (16S clone name, GenBank-linked accession number, BLAST-inferred identity); Macroscopic and Microscopic Image Data; and Student Collection Team Information. Using the administrative URL, all data can be entered and edited through restricted web-access. Using the public URL, users can view and query all data. Site Query results can be formatted to display any combination of geochemical parameters across one or more years and/or sites. Sequence Query results can be formatted to display inferred identity and GenBank-linked accession numbers across one or more sites. The RLMO Database, in its current form, is designed to accommodate physical, chemical, and molecular information as part of a five-year longitudinal study.

- Excerpted from TBI/Yellowstone meeting abstract (October, 2003)

In 2004, Will Drury expanded DNA Sequence data entry/archive screens, allowing us to enter information about DNA extraction procedures and PCR methods/primers utilized to generate each clone. Concomitantly, Will added appropriate sequence query boxes for searching and downloading this information. Secondly, Will added DGGE entry fields and we have begun archiving population genetics studies from long-term RLMO sites.

Sharing Information:

The RLMO database is provided on-line, as described above. Users can browse the public portion of this database in a non-restricted manner. A book chapter about this work, "The Red Layer Microbial Observatory Database: A Model for the Integration and Dissemination of Biological and Geochemical Data via the World Wide Web" is currently in press at the Thermal Institute of Biology, Montana State University.

Contributions

Contributions within Discipline:

In terms of molecular microbial population studies and systematics, the RLMO project continues to improve our understanding of GNS/Chloroflexi diversity. Ongoing studies suggest that temporally variable geochemical mechanisms drive site-specific selection, providing a model for bacterial evolution and selection. This year, we have greatly improved and streamlined our gel-based population genetics methods (DGGE). Specifically, we have used our RLMO database to develop and implement a battery of RLMO-specific controls (both phylum and sub-phylum level) for rapid identification of RLMO community members. These may be of value and interest to other researchers performing community assessments on comparable mat systems. In addition to assessing red layer community diversity, we have also begun applying population methods to neighboring/upstairs green Cyanobacterial layers in our efforts to understand the extent to which these two macroscopically distinct communities are microscopically separate (i.e. is there population mixing?). Our first study suggested that Red GNS/Chloroflexi are present in these Cyanobacterial layers and revealed extensive new bacterial diversity, some of which did not correspond to red layer-specific controls. Whether unique populations of consumers û e.g. Proteobacteria and/or Gram Positives - thrive in red vs. green layers is not clear at this time but has emerged as a new question. Whether specific Red GNS/Chloroflexi variants thrive in the green layer is also not clear at this time but is currently driving new hypothesis-testing.

The recently improved RLMO database (similar to Dr. Mary Ann Moran's SIMO Database), formally described in a book chapter (in press), continues to provide a model database for other PIs involved in long-term environmental monitoring studies to archive and analyze environmental and molecular data. In addition to helping us identify appropriate controls for population genetics/identification, we are finding it immensely useful for assessing and comparing long-term data.

In terms of undergraduate education, RLMO-based curricula has continued to provide exemplary models for comparable undergraduate courses. We presented these curricular approaches for teaching majors-level Microbiology (with an emphasis on culture-dependent and culture-independent approaches) and Computational Biology at ASM's General Meeting in May, 2004. Both were extremely well-received. We were thus surprised when our manuscript describing the latter course was rejected by ASM's Microbial Education journal for being too methods-heavy and assessment/impact-light. We have revised this manuscript for proposal submission to the National Science Teacher's 'Handbook of College Science Teaching: Theory, Research, and Practice.' In the mean time, we continue to maintain all these curricula plus the adjoining ASM poster on our RLMO website so that other insterested instructors can access them. We are also developing a new manuscript about the heavily-assessed/high impact majors-level Microbiology course for submission to ASM ME, unfortunately now reviewed only once a year (each October).

Contributions to Other Disciplines:

This project has impacted other disciplines in the form of valuable scientific data, adaptable curriculum models, and enhanced outreach/dissemination capabilities. In terms of scientific data, our now fully implemented geochemical monitoring and databasing efforts are providing non-molecular data that is of value, minimally, to ongoing thermal inventory studies by geologists in Yellowstone National Park. As stated elsewhere, most microbiologists who perform research in Yellowstone were highly motivated to enhance timepoint monitoring studies after sharing data with traditional geologists via the extremely useful and successful 'Geothermal Biology and Geochemistry in Yellowstone National Park' meeting (organized by the Thermal Biology Institute in October 2003).

In terms of curriculum, our Computational Biology course protocols provides highly adaptable models that can be tailored to many non-microbial systems (e.g. instead of a bacterial 16S project, use any number of eukaryotic markers to study evolution, diseases, development, etc.). In developing presentations and manuscripts about this work, such connections were emphasized heavily.

In terms of enhanced outreach/dissemination capabilities, this project had facilitated the launching of the extensively-described WAMS program (please see Activities and Findings). This program has provided an unprecedented number of new pre-college outreach venues for colleagues in Botany, Forensic Science, Anatomy/Physiology, Probability/Statistics, Marine Biology, and Geology.

Contributions to Human Resource Development:

In terms of providing opportunities for research and teaching in science and engineering areas, these topics have been extensively covered in the Participants and Activities sections.

In terms of improving access to and retention of underrepresented groups, we have been involved with the following: Through our partnership with Upward Bound (see Participants and Activities), we provided once-a-month pre-college activities for 4 local Hispanic females and 1 low-income/first-generation college-bound male. For our summer GERMS program, we selected 1 male and 5 female undergraduates from WOU (2 were Hispanic). Selection was based on merit- and experience-based criteria using an application form we developed. Unfortunately, we elected to turn down 1 Hispanic male and 1 African-American female from Ghana û among others û for this program. We have developed the new low-cost WAMS pre-college outreach program (see Participants and Activities) and are now able to offer need-based assistance scholarships and actively recruit low-income and underrepresented groups from the area. Finally, we have partnered with GEAR-UP (see Participants and Activities)à

Our development of new educational activities and providing assistance-based scholarships is exemplified best by our new WAMS program, described above and elsewhere.

Likewise, providing vehicles for exposing pre-college teachers and pre-college students to science and technology has been exhaustively covered in the Participants and Activities/Findings sections of this report.

Contributions to Resources for Research and Education:

These topics have been covered and summarized extensively in this report.

Contributions Beyond Science and Engineering:

Special Requirements

Special reporting requirements: None

Change in Objectives or Scope: None

Unobligated funds: less than 20 percent of current funds

Animal, Human Subjects, Biohazards: None

Categories for which nothing is reported:

Any Journal

Contributions: To Any Beyond Science and Engineering