## UNDERGRADUATE RESEARCH LONG TERM GRANT APPLICATION

### Budget Worksheet

<table>
<thead>
<tr>
<th>BUDGET ITEM</th>
<th>Department or College Funds</th>
<th>Outside Agency Funds</th>
<th>Personal Funds</th>
<th>Undergrad. Research Funds</th>
<th>GRAND TOTAL</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Materials</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Department of Botany:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disposables/other</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DNA Lab:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Purified Agarose</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>OUR:</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>RNA Isolation</td>
<td></td>
<td></td>
<td></td>
<td>$380.00</td>
<td></td>
</tr>
<tr>
<td>RNA Amplification</td>
<td></td>
<td></td>
<td></td>
<td>$350.00</td>
<td></td>
</tr>
<tr>
<td>Primers</td>
<td></td>
<td></td>
<td></td>
<td>$100.00</td>
<td></td>
</tr>
<tr>
<td>Sequencing</td>
<td></td>
<td></td>
<td></td>
<td>$725.00</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>$1955.00</td>
</tr>
<tr>
<td><strong>Equipment</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Research Scholarship</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(Max request $2,500.00)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Mileage to gather Data (.36 per mile)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>GRAND TOTAL</td>
<td>$400.00</td>
<td></td>
<td>$1555.00</td>
<td></td>
<td>$1955.00</td>
</tr>
</tbody>
</table>

### NOTES:

- Equipment and left-over materials purchased with this grant will remain the property of WSU.
- You may not request money for gas purchases for travel. WSU reimburses travel expenses at a set mileage rate only.
- Grant money cannot be used retroactively on previously existing expenses. Requests for reimbursements will be denied. All purchases must be made after receiving funding and clearance from the OUR office.
Analysis of genes for salinity tolerance in plants native to the Great Salt Lake ecosystem

Project Description

The salinization of arable land is an increasing problem throughout the world. Agricultural practices such as irrigation and land clearing lead to an accumulation of salts in soil. The effect is particularly prominent in arid environments. As the total amount of arid land continues to grow due to climate change, it will become necessary to use these saline soils to meet the agricultural needs of the world (Pitman and Läuchli, 2002). Plants use several different mechanisms to deal with salinity. Of these, the best understood mechanism is sodium exclusion. Salt tolerance is increased by the active transport of sodium ions out of the plant’s tissues by membrane bound proteins that act as pumps. The high-affinity potassium transporter (HKT) family of proteins is among the most studied groups responsible for contributing to salt tolerance via this sodium exclusion mechanism (Munns and Tester, 2008). Experimental evidence has shown that members of this family increase salt tolerance in genetically modified tobacco plants when compared to wild type plants (Chen et al. 2011).

The Great Salt Lake ecosystem is a natural source of diversity for salt tolerant plants (halophytes). Many native species of halophytes live on and around its shores in salt levels that would prevent the growth of other plants. The mechanisms for salt tolerance and the genes involved have not been studied in the majority of these species. While some investigation has been conducted on HKT genes in halophytes, most of the work has focused on agricultural species. In addition little work has been done investigating the HKTs of wild species in their natural habitat. For my project I propose to collect halophytes native to the Great Salt Lake ecosystem and analyze the messenger RNA (mRNAs) used to synthesize HKT proteins. A variety of halophyte species collected from the wild will be used as samples. The mRNAs coding HKT proteins will be isolated and amplified from root and leaf tissue using reverse-transcription PCR (RT-PCR), a simple and easy method which can be performed in any laboratory (Shao et al. 2008). Species which are successfully amplified will be sequenced. Once a sample has been selected and sequenced, the data will be used to construct a phylogenetic tree relating the sequence to other known sequences within the National Institutes of Health (NIH) and other online databases. It is my hope that this
project will help to further our understanding of these transporters and the role they play in sodium exclusion. Analysis of these naturally salt tolerant plants in their native environment may lead to new methods for dealing with the increase in soil salinity worldwide and the development of plants capable of coping with greater levels of salt within the soil.

I plan to work independently in my research. I will be responsible for gathering all samples and performing all the laboratory procedures. My mentor Dr. Harley will provide inspiration and insight when necessary. I will consult with my mentor before conducting any procedures. In addition I will be responsible for gathering and interpreting all of the data produced. Upon completion my final results will be reviewed. Any publication of my completed research will be under the guidance of my mentor.

<table>
<thead>
<tr>
<th>Dependent</th>
<th>Independent</th>
</tr>
</thead>
<tbody>
<tr>
<td>(Helping faculty do research)</td>
<td>(Student doing own research)</td>
</tr>
</tbody>
</table>

I am a Botany major and chemistry minor, working with an emphasis on genetics. I have taken several applicable courses including, Plant Genetics, Microbial Genetics, Molecular Genetics, and a full year of Biochemistry which included a course on laboratory techniques. My lab experience consists of work done in these and other courses. I have in addition worked independently in the DNA lab since 2011 where I have helped Dr. Jonathan Clark refine laboratory protocols. I have become proficient in many procedures including DNA isolation, PCR, and gel electrophoresis. I am familiar with all the protocols and techniques which will be used in my research. Overall I am extremely enthusiastic about this project and look forward to conducting the research. I am prepared to address any issues that may arise and I am committed to collecting accurate data for analysis.

Over the last summer I completed a directed thesis readings course with my mentor Dr. Harley which developed into this project. During this course I researched the background information needed to create my project. In my research I became familiar with the procedures necessary to perform my project and the relevant data already available. I have concluded based on the work of others that my project is both appropriate and feasible for my timeline and budget. I am now enrolled in a thesis research course in which I will conduct this project then write my thesis and present the results in a 45 minute presentation to an open audience. I plan to present my research project at the WSU Undergraduate Research Symposium and Celebration this coming spring.
and then submit my research for publication after it has been presented. In addition I plan to present my research at the meeting of The Botanical Society of America in Boise, this July 2014. I will also submit my sequence data to Genbank for inclusion in their online database.

**Project Methods**

This project uses standard laboratory techniques and equipment available in the WSU Science Labs. Native halophyte species in the Chenopodiaceae family will be used to isolate and analyze naturally occurring HKT genes. All samples will be collected from their native habitat around the northeastern shore of the Great Salt Lake. The GPS location of collection will be recorded for each sample. Specific species will be identified using standard morphological taxonomy and the Utah Flora: Chenopodiaceae key. RNA will be isolated from both the roots and leaves of each sample to test for expression in these tissues. The mRNAs coding HTK proteins will be amplified using RT-PCR. Samples which successfully amplify will be sequenced by a commercial laboratory. Sequence data will be analyzed using the NIH Genbank database. The MEGA software program and the HKT sequence data within the NIH database will be used to construct a phylogenetic tree to compare my transcripts to other known sequences.

**Timeline**

**Fall 2013:** Collect samples from around the Great Salt Lake; RNA isolation; HKT mRNA amplification; preliminary sequence analysis; Sequencing.

**Spring 2014:** Continue amplification and sequencing; Prepare data for analysis; Compare sequence to online database; Develop phylogenetic tree; Summarize research and present at WSU OUR Symposium; Write research paper.

**Summer 2014:** Present research at The Botanical Society of America; Prepare results for publication.
Budget Explanation:

The budget for materials is $1955.00. The department of Botany and the DNA lab are each providing $200.00 to cover the cost of incidentals and agarose gels. I am not requesting a research scholarship or travel expenses. The total amount of funding I am requesting from the OUR is $1555.00. This will cover the cost of an RNA isolation/purification kit, an RNA amplification kit, primers, and sequencing.

Laboratory Expenses:

<table>
<thead>
<tr>
<th>ITEM (Vendor)</th>
<th>COST</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trizol Plus RNA Purification Kit (Invitrogen)</td>
<td>$380.00</td>
</tr>
<tr>
<td>SuperScript III One-Step RT-PCR Kit (Invitrogen)</td>
<td>$350.00</td>
</tr>
<tr>
<td>Primers</td>
<td>$100.00</td>
</tr>
<tr>
<td>DNA Sequencing (Genewiz)</td>
<td>$725.00</td>
</tr>
</tbody>
</table>

References


1. What funding have you received from OUR in the past? Where has your previous project been disseminated?

This is my first project and I have never received funding from OUR in the past.

2. Is this project part of a required course? If so, please indicate the support (monetary and in-kind) provided for this project by the academic department.

No, my project is for an elective course, BTNY 4850 Thesis Research.

3. What additional sources of funding have been solicited? Is your department willing/able to fund any equipment they will be retaining?

The Department of Botany is supplying $200.00 for incidentals, and providing use of their laboratory and equipment. The Weber State DNA lab is supplying $200.00 to cover the cost of agarose gel electrophoresis.

4. Where do you plan to disseminate the results of this project?

I will present my research to an open audience upon completion in the Botany Department as part of my Thesis course. I will participate in the WSU Undergraduate Research Symposium and Celebration this next spring. I am also planning on presenting my research at the Botanical Society of America’s meeting in July 2014.

5. If you are requesting a Research Scholarship, please list all significant time commitments (5+ hours per week) that you expect to maintain over the duration of your project including, for example, class and work schedules.

I am not requesting a scholarship at this time.
Student Name (last, first): Wall, Ryan

Project Title: Analysis of genes for salinity tolerance in plants native to the Great Salt Lake ecosystem

Mentor Directions: After carefully reviewing the proposal and assessing both the viability of this project and the qualifications of the student requesting funding, answer the questions found below. Please expand the sections as necessary (do not attach separate letter). If the project involves the use of human subjects or protected animals, be sure the student secures IRB or ACUC approval. If the project receives funding, it is your responsibility to work closely with the student, monitor the ongoing progress of the project and budget, and evaluate the project’s results. Failure to do so will jeopardize funding for this project and any future projects.

1. How long and in what capacity have you known this student?

I first met Ryan when he and was in my Plant Form and Function (BTNY 2104) class in Spring 2010. Since then I have had Ryan as a student in Biology of the Plant Cell (BTNY 3153) and Plant Genetics (BTNY 3303). The idea for Ryan’s project came from his desire to do a molecular biology-based thesis project. I told him that if he could come up with a research question that could be answered by using molecular techniques, then I would be his mentor. Inspired by the plants around the Great Salt Lake, he decided to investigate mechanisms of salt tolerance. He planned his research project during the Summer 2013 semester to fulfill the requirements for BTNY 4840, Thesis Readings, with me. He is now trying to get the funding to do his planned project so that he can complete BTNY 4850, Thesis Research.

2. Briefly describe the proposed project. Is this part of a larger research project? Is this part of a course? If so, how is the project apart from the nature and scope of activities normally taken for the course (Please attach a copy of your course syllabus)?

Ryan’s proposed project fulfills the requirements for a thesis in Botany. For the various steps of the thesis process, he will get a total of 6 credit hours in three independent study classes, BTNY 4840, 4950, and 4970.

3. Give an assessment of the project’s significance to the student’s discipline and of the project’s educational and/or professional benefit to the student.

This project gives Ryan experience with planning, conducting, and reporting on an independent research project in preparation for graduate school or professional employment in plant molecular biology. It will also enable him to integrate techniques that he has learned in a variety of courses in Botany, Chemistry, and Zoology.

4. Comment on the qualifications of the student to successfully complete this project, both in terms of the project’s scope and its time frame.

Ryan is well qualified to complete this project. His courses in Botany, Chemistry, and Zoology as well as the work that he has done in the WSU DNA Lab have given him the skills needed to complete the field and lab portions of his project as well as to analyze his data in his proposed time frame.

5. Comment on the justification and appropriateness of the project budget, including the necessity of a stipend (if requesting one).
The project budget is appropriate. The two main expenses are kits and contracted work. The kits greatly simplify the work needed to extract and amplify the genes that he will be investigating. Because of equipment costs, contract labs are increasingly used to do the work of synthesizing primers and sequencing DNA. These contract labs make it possible for institutions like WSU to engage in the type of molecular biology investigation that Ryan is proposing.

6. Describe your role in the project.

My role is to supervise and evaluate all steps of Ryan’s thesis. I serve as a sounding board for ideas, editor, technique instructor when needed, field work buddy, and sometimes lab assistant (because sometimes you just need an extra pair of hands). The project idea, development, and execution are all Ryan’s.

7. Include anything else that you think will be helpful to the committee in evaluating this application.

This project ___ DOES XX ___ DOES NOT require review by the WSU Institutional Review Board for Human Subjects or the WSU Animal Care and Use Committee.

[Signature]
Project Mentor Signature

[Date]

2504
Campus Mail Code

7434
Phone Extension