**Undergraduate Research Semester/Exploratory 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>Materials</td>
<td>- DNAeasy DNA extraction kits</td>
<td></td>
<td></td>
<td>- Sequence generation from PCR products ($14/sample) for 65 samples</td>
<td></td>
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<td></td>
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<td></td>
<td></td>
<td>Total = $1,600</td>
<td>$2,510</td>
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<td></td>
<td>- QiaQuick PCR purification kits</td>
<td></td>
<td></td>
<td>Total = $910</td>
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<td></td>
<td>- Material to perform PCR reactions</td>
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<tr>
<td></td>
<td>Total = $1,600</td>
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<tr>
<td>Equipment</td>
<td></td>
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<tr>
<td>Mileage to gather Data (.36 per mile)</td>
<td></td>
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<tr>
<td>GRAND TOTAL</td>
<td>$1,600- Dr. Marshall’s start-up funds</td>
<td></td>
<td></td>
<td>$910- requested from OUR</td>
<td></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.
Investigating a Potential Hybrid Zone in Fence Lizards

Project Description

Utah is home to four different species of Spiny lizards (genus Sceloporus). In southwestern Utah (Washington County) all four species come into close proximity with each other. The distribution of two of these species the Eastern Fence lizard (Sceloporus undulatus) and the Western Fence lizard (Sceloporus occidentalis) overlap in several localities in the Pine Valley Mountain area of Washington County. This is the only region in the world that these two species meet. Based on morphological observations several employees (personal communication with Dr. Mark Grover) of the Utah Division of Wildlife Resources (DWR) believe these two species of lizards are occasionally breeding and forming hybrid offspring. The extent or even existence of this hybrid zone needs to be confirmed through mtDNA sequencing and analysis.

Hybrid zone dynamics have been found and studied at a more shallow level between major lineages within the Undulatus species group (Leache and Cole, 2007) but not at the deeper level described above. This pilot project could provide concrete identification of the hybrid zone as well as foundational work for further investigation into hybrid zone dynamics, selection, species boundaries, and speciation studies. It has been noted in previous research, that due to their wide geographic distribution, spiny lizards are considered model species in conservation biology (Angilletta et al. 2004).

In this study, I will be using the mitochondrial ND1 gene to assess evidence of hybridization resulting in gene flow between species. The mitochondrial ND1 gene has been used to assess data in various spiny lizard genetic studies, including Leache and Reeder (2002). To establish baseline genetic markers (haplotypes) typical of both S. occidentalis and S. undulatus, 50 sample sequences were downloaded from GenBank (www.ncbi.nlm.nih.gov/genbank) to create a reference sequence library. In addition to the above sequences, at total of 20 reference tissue samples were donated from sites within Utah
but distant to the potential hybrid zone location (Dr. Mark Grover) and from various localities in
the Mojave desert (PhD. student Dean Leavitt, San Diego State University).

During the summers of 2010 and 2011 lizard tail samples were collected from 45 lizards
morphologically identified as either *S. occidentalis* or *S. undulatus*. These tissue samples were
taken from four different potential hybrid zone localities in the Pine Valley mountain area. These
tissues are being stored in the -20° C freezer in the College of Science DNA sequencing facility.

This research grant requests funds for laboratory materials to generate PCR products and
fees required to send PCR products to a private facility (GeneWiz) for sequencing. Once
samples are sequencing, DNA editing and analysis will be done at the DNA sequence analysis
station in the College of Science DNA sequencing facility.

*My Role*- In the lab I will be responsible for generating mitochondrial DNA PCR products
from samples of both lizard species. I will independently generate PCR products, sequence
data, analyze data, and generate hybrid zone map localities. Throughout this project I will
continue to learn lab techniques and develop into an independent researcher.

Dependent____________________________ X Independent

*Previous Training and Experience*- I am working towards a bachelor of Integrated Studies
Degree with emphasis in Zoology/Botany/Environmental Studies with my graduation date set for
April of 2012. I have helped organize the herpetology collections at Weber State University. I
have volunteered on numerous types of fieldwork and gained hands on experience with various
reptiles and amphibians. I have also been employed on a number of biology related jobs,
including black bear, mountain lion, and deer projects. I also have a considerable amount of
lab experience. My job duties have consisted of capturing specimens, collected DNA samples,
taking measurements, and analyzing data.
I have had the opportunity to work with Dr. Marshall since April of 2011. I look forward to learning more from him and continuing to build my knowledge and understanding of biology research. Throughout the course of 2011 I have continued to improve in all aspects of research. I now have a solid foundation for collecting, analyzing, and presenting data, as well as reading and understanding important information.

This opportunity is especially important to me, because I plan on attending a research based graduate program in 2013. This experience will be very important in gaining acceptance into a program as well as aiding in my future success. I am passionate about this work and I am excited to continue to learn and grow as a researcher.

**Product** - I plan to present this research in August of 2012 at the World Herpetology Congress in Vancouver, Canada. ([http://www.worldcongressofherpetology.org/](http://www.worldcongressofherpetology.org/)).

**Project Methods & Timeline**

I will be analyzing 45 tail samples collected from both *S. occidentalis* and *S. undulatus*. These samples were taken from four different potential hybrid zones in the Pine Valley mountain area. I will extract DNA from these 45 samples using a DNeasy blood and tissue kit. I will also be using 50 baseline samples from GenBank and 30 donated tissue samples for comparison. These samples were taken from areas distant to the potential hybrid zone area.

I will sequence the entire mitochondrial ND1 protein coding gene (969 bp) and using lineage specific PCR primers (SnowF and SnowR). I will do this following the protocols found in Leache and Cole (2007). In that paper approximately 10–25 ng of total DNA was used as template for PCR in a final volume of 25 μL containing 1× buffer, 1.5 mm MgCl2, 0.1 mm of each dNTP, 0.5 μm of each primer, and 1.25 U of Taq polymerase. Sufficient PCR product for direct sequencing will be generated after 36 cycles (94 °C for 30 s, 58 °C for 1 min, 72 °C for 1 min). PCR products will be purified using QIAGEN PCR purification kits (QIAGEN Inc.). Work will be done in
the Weber State College of Science DNA editing and analysis station. PCR products will be sent to GeneWiz Inc. (South Plainfield, NJ) for sequencing.

**Timeline**- I plan to work throughout the spring semester of 2012 and into the summer of 2012 with Dr. Marshall. My goal is to have a general abstract completed by March of 2012 for the World Herpetology Congress. I then plan to have finished analyzing the data by July of 2012.

**Budget Explanation**

Dr. Marshall has start-up funds that he will be using to help fund this project. The $1,600 required for DNA extraction from tissue, PCR reactions, and purification of PCR products will come from these funds. I am requesting $910 to fund the sequence generation from PCR products ($14/sample) for 65 samples. These funds will allow me to finish this initial phase of the research project and be in a position to have data generated an provisional conclusion to present a poster at the World Herpetology Congress this upcoming summer.

**Literature Cited:**


Additional Questions

1. What funding have you received from OUR in the past, Where has your previous project been disseminated. *I have not received any previous funding.*

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, it is not required by a specific course.*

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

   *Funds from Dr. Marshall’s Start-up fund will be used. N/A*

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

   *World Congress of Herpetology, August 8-14 of 2012*

5. If you are requesting a stipend, 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. *N/A*
UNDERGRADUATE RESEARCH SEMESTER/EXPLORATORY GRANT

APPLICATION

FACULTY MENTOR RECOMMENDATION FORM

Student Name (last, first): Shaney, Kyle _________________________                  ______

Project Title: Investigating a Potential Hybrid Zone in Fence Lizards _________________________

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? 2 years

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)? Kyle’s proposed project will be the culminating part of this on-going project. Kyle has been part of the field component of this project. He traveled to Southern Utah last year with me to help with sample collection. I am trying to give Kyle some experience in what a mixed field/DNA lab project would be like for graduate school. His proposed project would help fund the lab part of his experience.

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. Kyle has a strong desire to go to graduate school. However, Kyle struggles in the classroom but is very good out in the field collecting and identifying amphibians and reptiles. He is also much more of a hands-on, apprentice style learning. Kyle’s involvement in research is where he is getting a good share of his education. If Kyle can complete this project and present it at the World Herpetology Congress this summer that will give him an opportunity to meet with and discuss potential Masters projects with researchers at this conference. In my opinion, without this kind of conference interaction with a potential mentor Kyle will have a difficult time getting into graduate school.

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. In my opinion I think this project can be accomplished in this time frame. Furthermore, as mentioned above, the student should be sufficiently motivated to accomplish it in time to present at the meetings this summer.

5. Comment on the justification and appropriateness of the project budget, including the necessity of a stipend (if requesting one). In my opinion Kyle’s budget is appropriate given the shared resource contributions.
6. Describe your role in the project. *I will be heavily involved in mentoring Kyle. Kyle has some experience from the laboratory component of his Genetics courses but I will still need to provide significant mentoring in the laboratory and analysis portions of this project.*

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

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

___ Jon Marshall 1/27/12  
Project Mentor Signature Date

2505  
Campus Mail Code Phone Extension