

## UNDERGRADUATE RESEARCH LONG TERM GRANT APPLICATION Budget Worksheet

BUDGET ITEM ✓	Department or College Funds	Outside Agency Funds	Personal Funds	Undergrad. Research Funds	GRAND TOTAL
Materials		-Slides, slide boxes, antibodies, regents (3k) *			
Equipment	-Cryostat (15 k) - Ultracold freezer (8k)	-Microscope with camera (15k) *			
Stipend: Hrs @ \$10/hr Benefits @ 8.5% Total				250 hrs * \$10/hr = <u>\$2,500 +</u> 8.5% = <u>\$2,712.50</u>	\$2,712.50
Mileage to gather Data (.36 per mile)		-72 mi per trip (\$26) Est 10 trips \$260.00 *			
GRAND TOTAL				2712.50	\$2,712.50

### 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.

Students do not receive "benefits" money.

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\*Dr. Meyers' NIH grant covers the cost of travel to the University of Utah for picking up tissue, as well as all consumable chemicals and supplies (e.g., antibodies, microscope slides, chemical reagents, etc.). In addition, the Zoology Department currently has all of the equipment needed for our work, such as the cryostat and the ultracold freezer.

## UNDERGRADUATE RESEARCH LONG TERM GRANT APPLICATION

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### **Sexual Dimorphism Of Syringeal Muscles In Female And Male Songbirds**

**Description:** The syrinx is a complex vocal organ found in birds at the tracheobronchial junction (Suthers et al. 1999) and is made up of four pairs of muscles, dorsal and ventral tracheobronchialis and syringealis. Previous work has shown that the syrinx is made up of two muscle fiber types, fast and superfast (Meyers et al. 2005). These superfast fibers have been shown to contract at 250 Hz (Elemans et al. 2008), and are an uncommonly evolved characteristic found in sound producing organs of some fish and in the rattlesnake tail shaker muscle.

Syringeal muscles have been the focus of our laboratory's research over the past several years. We have investigated the syringeal muscle fibers of various songbird species and have quantified and compared the ratios of superfast fibers between males and females. In Zebra Finches and Bengalese Finches, females do not sing, and males were found to have significantly more superfast muscle fibers (70% vs. 30% and 85% vs. 25%, respectively). In European Starlings, where males and females sing, both sexes displayed similar percentages of superfast fibers (~70%). These results are unsurprising since superfast fibers are attributed to singing. However, Brown-headed Cowbirds showed similar percentages of superfast fibers (67%) between males and females, in spite of the fact that the females do not sing. This suggests that superfast fibers may be characteristic of the songbird syrinx, rather than attributed to sexual differences in singing (Uchida et al 2008).

Over the past summer we have collected data on a two different species such as Yellow-headed Blackbirds and Brewer's sparrows. Yellow-headed blackbirds were of particular interest to us, as they are in the same family as Brown-headed Cowbirds. In the Blackbirds, in which both males and females sing we discovered similar ratios of superfast fibers between the two genders (~59%). In Brewer's Sparrows, where males sing and females do not, we also found similar ratios between the two genders (~71%). My goal for the next couple of semesters is to expand our summer findings by

examining additional species (e.g. Canaries, Brewer's Blackbird, Red-winged Sparrows, White-crowned Sparrows), as they can be obtained. I would also like to get additional individuals of Yellow-headed Blackbirds and Brewer's Sparrow, in order to increase our sample size.

Our long term goal is to look at a broader range of birds in different families. Birds of great interest would be more species in the family of the finches and the cowbirds. Hopefully this will give us greater insight into why the finches show significant sexual dimorphism in superfast fiber type or why non-singing females, such as cowbirds and sparrows, have comparable percentages with their singing counter parts. We are also interested in looking at Western Kingbirds, since they are suboscine and can be considered an out-group.

I will analyze and quantify individual fiber types (fast, superfast) within the syrinx of different species as they become available, or as they are raised by Dr. Franz Goller, our collaborator, at the University of Utah. Ultimately, I aim to determine the function and explanation of presence of these superfast fibers.

**Role:** In the lab I will be responsible for performing immune-histo chemical reactions and photographing the results; I also will be generating a photomontage of syringeal muscles that can then be used to measure fiber diameters and percentages. Over the summer I progressed from being dependent to independent in our research. I have become competent and confident in all of our routine laboratory procedures. I will also be involved in training new students that have come into the lab.

Dependent \_\_\_\_\_  \_\_\_\_\_ Independent  
(student helping faculty do research)                      (student doing own research)

**Previous training and experience:** I am working towards a Bachelors of Science degree in Zoology, with a minor in chemistry and with a strong mathematic and microbiology influence. I have completed the introductory classes and multiple upper division classes including Anatomy, Physiology, and Histology. Next semester I will be finishing up with Biochemistry, Comparative Physiology, and other supporting classes.

I began to working with Dr. Meyers in February 2009, and I am very eager to keep learning more from him and build my research experience. Over the summer I have learned and progressed in my abilities including: improving critical thinking skills, laboratory procedures, analyzing data, reading scientific papers, and presenting the collected data. I have also be strengthened my knowledge in histology, chemistry, anatomy, and physiology; all of which will assist me in my future experimentation in this lab.

This opportunity is important to me given that I plan to enter a research-based graduate program; I know this experience will aid in my acceptance into, as well as my success in one of these programs.

**Product:** We presented this preliminary data at the Utah Ornithological Society meeting in August of 2009. I plan to present this research in the spring of 2010 at the WSU Undergraduate Research Symposium and Celebration. We also expect to present it at the Society of Integrative and Comparative Biology (SICB) meeting January 2011 in Salt Lake City.

#### Project Methods & Timeline

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(Approximately 1 page)

**Methods:** The muscle tissue will be collected with Dr. Franz Goller, at the University of Utah. The tissue will be frozen at  $-150^{\circ}\text{C}$  in liquid nitrogen and stored in an ultracold freezer at  $-80^{\circ}\text{C}$ . The tissue will then be cut on a freezing microtome into  $10\ \mu\text{m}$  slices and placed on microscope slides. Our lab routinely uses antibodies and myosin ATPase histochemistry to identify muscle fiber types as described in Meyers and Mathias (1997) and McFarland and Meyers (2008). All reactions will be photographed, counted, measured, and quantified for interpretation.

**Timeline:** I plan to work over the length of the Fall 2009 and Spring 2010 semesters with Dr. Meyers and Dr. Goller. My goal is to have enough information to write and abstract for the 2011 SICB meeting by the end the school year.

Dr. Meyers has been awarded an NIH grant that finances the equipment and chemicals used in our laboratory. Over the next two semesters I plan to complete one experiments per week which include tissue collection, histological runs, photography, and analysis of results. Each experiment takes about 15 hours to complete, so 15 hr per week for 30 weeks equals 450 hours. I am requesting the amount of \$2,500, as a stipend for the time I will devote to working in the lab. Since devoting this much of my time to the lab I will not be able to work enough at my other job to supplement my income. As a result, I will be relying on the research with Dr. Meyers as major source of income.

#### Literature Cited

- Elemans, C.P.H., A.F. Mead, L.C.Rome, and F.Goller.2008. Superfast vocal muscles control song production in songbirds. *PLoS ONE* 3(7):e2581.doi 10.1371/ journal.pone.0002581.
- McFarland, J.C., and R.A. Meyers. 2008. Anatomy and histochemistry of hindlimb flight posture in birds.1. The extended hindlimb posture of shorebirds. *Journal of Morphology* 269(8):967-979.
- Meyers, R.A., and E. Mathias. 1997. Anatomy and histochemistry of spread-wing posture in birds 2. Gliding flight in the California Gull, *Larus californicus*: A paradox of fast fibers and posture. *Journal of Morphology* 233:237-247.
- Meyers, R.A., D. Schmutz, and F. Goller.2005. Histochemical analysis of songbird syringeal muscles. *Integrative and Comparative Biology* 45(6): 1167
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- Uchida, A.M., Green, J., Ahmad, S., Goller, F. and R.A. Meyers. 2009. Sexual dimorphism of syringeal muscles in songbirds. *Integr. Comp. Biol.* 49 (1): e318.