Office of Research Summer Workshop Series
Crafting a Competitive Proposal
July 2017

COLOR KEY

Hook
What is Known
Gap in Knowledge
Critical Need

Long-Term Goal
Hypothesis and Proposal Objectives
Rationale
Qualifications

Innovation
Expected Outcomes/Deliverables
Impact

NIH SPECIFIC AIMS PAGE

1st Paragraph

Hook

- Why should we care?
- Establish a sense of importance or urgency.

What is Known

- Provide background info necessary to understand why you are proposing the work.
- KEY DETAILS ONLY.

Gap in Knowledge

- Piece of information that is not known.

Critical Need

- What is the knowledge that you propose to develop?
- Why is this the logical next step for advancing the field?

2nd Paragraph

This paragraph should answer these questions:

- *What do you want to do?*
- *Why are you doing it?*
- *How do you want to do it?*

Long-Term Goal

- Must align with funder's objectives
    - Research
    - Should remain general

Hypothesis and Proposal Objectives

- You want to demonstrate to the reviewers that you have a hypothesis-driven proposal that is testable.
- Describe how your project addresses the critical need.
- Clearly state the proposed solution.

Rationale

- Explain how you arrived at your central hypothesis (past studies, published literature, etc.)
- State what your project's completion would make possible and TIE IT TO THE FUNDING AGENCY'S MISSION

Qualifications

Specific Aims

3rd Paragraph

Innovation

Expected Outcomes/Deliverables

Impact

Specific Aims

https://www.niaid.nih.gov/grants-contracts/draft-specific-aims

https://www.niaid.nih.gov/grants-contracts/sample-applications

- Avoid Aims that depend on only one outcome
  - ~~"Does A cause B?"~~
  - "Does A cause B or non-B?"
- Avoid Aims that are primarily descriptive – this belongs in preliminary data
  - "We will measure levels of X in 1,000 samples of Y to characterize the pattern of expression of X"

Iterative Process for Proposal Preparation (from NIAID website):

1. Staying in your niche, propose a project that
   - Addresses a highly significant problem
   - Is innovative—can create new knowledge
2. Outline draft Specific Aims and one or more hypotheses.
3. Identify a potential funding institute and a study section that would likely embrace your research.
4. Outline experiments.
5. Assess feasibility.
6. See whether you have access to all needed resources and expertise.
7. Make sure the project is not growing too big for your targeted time and budget.
8. If you hit a roadblock, go back to the failure point and revise your plans.

HYPOTHESIS AND SPECIFIC AIMS:

The transcription factor FOXP3 is critical to the regulation of numerous debilitating human immune-mediated diseases, the prevalence of which together affect over 8.5 million people (1 in 31 U.S. residents). In Inflammatory Bowel Disease (IBD) chronic intestinal inflammation indicates aberrant *in vivo* FOXP3+ T regulatory (Treg) cell function (1). Similarly, proinflammatory signals *in vitro* impair Treg function (2). Our lab was the first to characterize the essential role for the histone methyltransferase (HMT) EZH2 in the epigenetic regulation of FOXP3 (3). Recent published work extended our observations indicating a key role for EZH2 in FOXP3 repressor function (4); however the regulation and biological impact of the FOXP3-EZH2 pathway to IBD is unknown. This knowledge is important given the apparent loss of function of Treg cells in inflammation.

Our *long-term goal* is to dissect epigenetic mechanisms regulating Treg cellular differentiation and function, particularly within the setting of GI inflammatory diseases; as these discoveries will facilitate design of human cell therapy trials for IBD. Consequently, the *objective* of this grant is to characterize the role for the epigenetic regulator EZH2 in Treg suppressive function. These investigations are strongly supported by preliminary data demonstrating that: 1) EZH2 is required for Treg suppressive function; 2) IL6 signaling leads to phosphorylation and inhibition of EZH2; 3) lymphocytes isolated from the intestine of IBD patients demonstrate activation of IL6-induced gene networks and loss of EZH2 HMT function; and 4) conditional knockout of EZH2 in FOXP3+ T cells leads to *in vivo* immune dysfunction. Based upon these compelling data we propose the **CENTRAL HYPOTHESIS that EZH2 plays a critical role in the homeostasis of Treg cells, and the disruption of EZH2 function by inflammatory signaling pathways contributes to IBD.** Our rationale is that identification of the mechanism(s) to restore Treg suppressive function in the setting of intestinal inflammation will offer new therapeutic opportunities within the field of IBD. Our specific aims will test the following hypotheses:

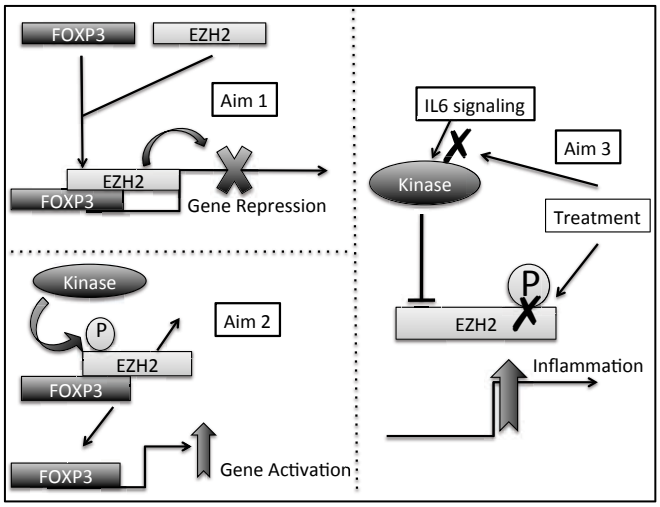

Figure 1: Conceptual framework. Through the mechanistic experiments designed in the following aims we will identify the role for FOXP3 in the recruitment of EZH2 to core target genes required for Treg function (Aim 1). We will define the signaling network responsible for phosphorylation of EZH2 and disrupted HMT function (Aim 2). Finally, we will perform pre-clinical studies of innovative therapy designed to generate Treg cells resistant to disruptive modifications in the setting of inflammation (Aim 3).

Aim 1: Repression of immunoregulatory gene networks by FOXP3 requires the formation of a complex between this transcription factor and EZH2.

Aim 2: Inflammatory stimuli, such as IL6 lead to EZH2 phosphorylation and thereby disrupt the enzymatic activity of this epigenomic regulator.

Aim 3: Inhibition of the IL6 to EZH2 signaling pathway permits sustained Treg suppressive function in the setting of intestinal inflammation.

Upon conclusion, we will understand the role for EZH2 in Treg loss of function in the setting of active inflammation. This discovery will stimulate new areas for experimental therapeutics in human chronic inflammatory diseases. Our environment in the Epigenetic and Chromatin Dynamics Laboratory combined with the Department of Immunology at the Mayo Clinic makes us uniquely qualified to pursue this objective given the extensive collective experience of histone methyltransferase biology, proinflammatory signaling networks, and *FOXP3* gene regulation.

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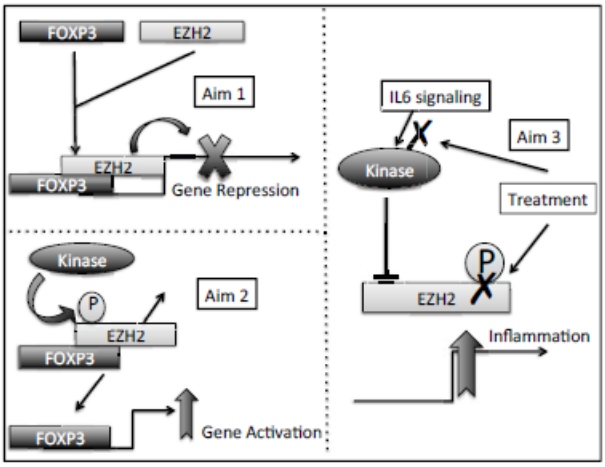

Figure 1: Conceptual framework. Through the mechanistic experiments designed in the following aims we will identify the role for FOXP3 in the recruitment of EZH2 to core target genes required for Treg function (Aim 1). We will define the signaling network responsible for phosphorylation of EZH2 and disrupted HMT function (Aim 2). Finally, we will perform pre-clinical studies of innovative therapy designed to generate Treg cells resistant to disruptive modifications in the setting of inflammation (Aim 3).

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PROJECT SUMMARY/ABSTRACT:

The transcription factor FOXP3 is critical to the regulation of numerous debilitating human immune-mediated diseases. Very recently, the essential role for the histone methyltransferase (HMT) EZH2 in the epigenetic regulation and function of FOXP3 has been described. Inflammatory pathways modify EZH2 activity, and inflammatory signaling impairs Treg function *in vivo* and *in vitro*. The biological impact of the FOXP3-EZH2 pathway to IBD is unknown. Our *long-term goal* is to dissect epigenetic mechanisms regulating Treg cellular differentiation and function, particularly within the setting of GI inflammatory diseases. These discoveries will facilitate design of human cell therapy trials for IBD. The *objective* of this grant is to characterize the role for EZH2 in Treg suppressive function. The *central hypothesis* is that EZH2 plays a critical role in the homeostasis of Treg cells, and the disruption of EZH2 function by inflammatory signaling pathways contributes to IBD. Our *rationale* is that identification of the mechanism(s) to restore Treg suppressive function in the setting of intestinal inflammation will offer new therapeutic opportunities. Our *specific aims* will test the following hypotheses: (Aim1) Repression of immunoregulatory gene networks by FOXP3 requires the formation of a complex between this transcription factor and EZH2; (Aim 2) Inflammatory stimuli, such as IL6 lead to EZH2 phosphorylation and thereby disrupt the enzymatic activity of this epigenomic regulator; (Aim 3) Inhibition of the IL6 to EZH2 signaling pathway permits sustained Treg suppressive function in the setting of intestinal inflammation. Upon conclusion, we will understand the role for EZH2 in Treg loss of function in the setting of active inflammation. *This contribution is significant* since it will establish that several pathways targeted by available therapies (ie IL1β, IL6, TNFα) have the potential to regulate EZH2 HMT activity through post-translational modifications. Furthermore, current Treg cell therapy trials, while promising have not addressed the key issue of *in vivo* inflammation-induced disruption of Treg function. *The proposed research is innovative* because we investigate the effect of inflammatory signaling pathways on epigenetic complexes in Treg cells, a heretofore-unexamined process. Insight into epigenetic mechanisms is impactful as T cell progenitor cells inherit the parent transcriptional profile and unlike genetic change, they are modifiable by currently available therapy.

NEA ARTWORKS SUMMARY

Project Description:

Ornamental needlework, especially American samplers, has long been a staple component of American "folk art." However, throughout most of the 20th century, Southern examples were thought to be exceedingly rare. With the advent of widely available online tools for genealogical research combined with a determination by institutions to exhibit and interpret these important works of art, scores of Southern examples have been identified and added to the American canon, though Georgia works have continued to be rare and have not received adequate attention. Girlhood Needlework in Colonial and Antebellum Georgia is the first comprehensive exhibition and publication of samplers made in the state during the late-18th and first half of the 19th centuries. Organized by the Georgia Museum of Art, the project will promote and disseminate original scholarship, and feature programs for general and academic audiences.

Major Project Activities:

The Green Center has tracked a number of Georgia samplers and approximately 50 of certain Georgia origin can be identified. Dale L. Couch, curator of decorative arts at GMOA, and Kathleen Staples, textile conservator and noted textile historian, propose to exhibit 25-35 samplers of Georgia origin and interpret their physical and documentary evidence contextualizing these compelling works of art.

Needlework in general is connected to female craft and samplers are most often connected to girlhood education, a topic that takes on unique dimensions in Georgia history. Couch will write an essay on a young sampler maker from Putnam County and trace her life through the records of her father, brother and husband in an effort to demonstrate methods of documentation for the lives of marginalized individuals. Her sampler will be included in the exhibition. Jenny Garwood of the Museum of Early Southern Decorative Arts (MESDA) will write on the sampler by Mary Smallwood from the MESDA collection.

Kathleen Staples will identify and describe the technical aspects of the works and she will write a broad interpretative essay on their social historical context, addressing such issues as girlhood education, stylistic influences and ethnic clues embedded in these stitched objects.

The book will include a chronology of Georgia, bibliography, foreword, appendices, and images of each work in the exhibition. GMOA will design and produce a rack card featuring a key to the embroidery stitches in the samplers.

The exhibition will be displayed in GMOA's Knox II Gallery, designed and installed in order to highlight the fragile and delicate works of art. Staples and Couch will give public tours of the exhibition throughout its four-month run, as will docents. Prior to the opening GMOA will host a Lunch & Learn on the exhibition for UGA faculty to open opportunities for collaboration. A Family Day will include gallery activities that facilitate interaction between children and works of art and a studio component in which participants create their own needlework using materials and techniques derived from the exhibition. The Senior Citizens Outreach will focus on Girlhood Needlework, with an audience from local senior centers with a particular interest in needlework and quilting.

UGA will be the site of the 2016 MESDA Textile Seminar, cosponsored by GMOA and the UGA Special Collections Library. Girlhood Needlework will be the highlighted exhibition; GMOA will host the keynote lecture, opening reception, and a gallery talk by the curators.

Girlhood Needlework will be shown concurrently with exhibitions of Cherokee baskets and recent acquisitions in the decorative arts during the 2016 Henry D. Green Symposium of the Decorative Arts, where Ms. Garwood will lecture on Georgia textiles before 1860. GMOA will publish her paper with the complete symposium proceedings, a post-project activity.

NSF PROJECT SUMMARY

Overview

"The overview includes a description of the activity that would result if the proposal were funded and a statement of objectives and methods to be employed."

Hook
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Intellectual Merit

"The statement on intellectual merit should describe the potential of the proposed activity to advance knowledge."

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Innovation

Broader Impacts

"The statement on broader impacts should describe the potential of the proposed activity to benefit society and contribute to the achievement of specific, desired societal outcomes."

Impact

Address how the research will promote teaching, training, research, and learning.

Describe plans to advance the participation of underrepresented groups.

Describe how science and technology will be enhanced.

PROJECT SUMMARY

Overview:

OVERVIEW: The overarching objective of this project is to elucidate the ecological, biochemical and molecular processes impinging on the evolution of diverse light spectra in the sexual signals of fireflies. The proposed studies will investigate the molecular basis of firefly spectral phenotype and selection by addressing the three components of a new integrative model for the evolutionary diversification of firefly mating communication: (1) The light produced by the enzyme luciferase has an emission spectrum that is directly controlled by its amino acid sequence; (2) This luciferase spectrum is altered by light-filtering screening pigments located in the photocytes of the light organ; (3) The resulting firefly light spectrum (interaction of luciferase and screening pigments) is subject to selection for optimal contrast with the ambient light environment during signaling. We will test and tune our model through a combination of fieldwork (e.g. ambient light conditions and firefly light spectra) and molecular analyses (e.g. sequencing, expression studies and mass spectrometry). Integration of all these data in our model will provide a significant advance in understanding the diverse bioluminescence spectra within and among firefly species. Moreover, this work will provide an unprecedented functional view of the molecular evolution of sexual signaling, and will directly inform future research that will connect phenotypic variation and fitness (e.g. field experiments on signal detection and the possible impact on mate attraction and mating success).

Intellectual Merit:

INTELLECTUAL MERIT: Comprehensive understanding of adaptation is a primary goal of evolutionary biology. However, knowledge of the relationship between genetic variation at the molecular level and variation in a particular adaptive phenotype is often limited. This represents an important knowledge gap and prevents a complete understanding of the evolution of sexual communication. Fireflies provide one of the few research systems where it is possible to rigorously link molecular variation, sexual signal phenotypes, and environmental variables to determine the molecular basis of selection and provide the knowledge base for a comprehensive analysis of signal evolution. The proposed research investigates the evolution of bioluminescence spectra in fireflies, a beetle family (Lampyridae) with more than 2000 species worldwide. The species-rich Photinus genus includes 34 species in North America. The intensity peaks in their bioluminescence spectra range from 555 to 580nm, which appear green to yellow/orange to the human eye. The Photinus study system has an established phylogeny and is ideal for studying molecular evolution and connected phenotypes because of the documented variation in bioluminescence spectra, DNA and AA sequences of luciferase enzymes, onset of daily activity (ambient light conditions), and habitat types. This research will substantially advance the understanding of interactions between genetic and environmental factors influencing the molecular basis of visual signal production and signal phenotypes, and ultimately the evolution of visual communication systems.

Broader Impacts:

BROADER IMPACTS: The project includes important components to capitalize on the broad public fascination and familiarity with fireflies to engage STEM learners. Undergraduate and graduate students will work with the Luft group to develop outreach activities around fireflies that integrate research findings and educational value. These activities will be part of Street Corner Science, which brings science and mathematics activities to under-represented and under-served students in the community during the summer months. (80% of K-12 students in the Athens Clarke County School District qualify for free lunch and 71% are minority.) All developed materials will be made available online to outreach personnel across the US. In addition, we will specifically recruit women as well as under-represented minority students (through the Peach State Louis Stokes Alliance for Minority Participation program at the University of Georgia) to participate in all aspects of our research program.

CAREER: Understanding the Basis of Maize-Microbiome Interactions

Project Summary

Overview

All land plants have trillions of microorganisms living on, around, and inside them. These communities are collectively called a plant's "microbiome." Most of our knowledge of plant-microbe interaction is limited to pathogens and a few specific symbioses. Little is known about the forces that shape the larger microbial community and the effect that this community has on plant health and performance.

The overarching goal of this project is to identify and model the influences that act on different microbial communities living on and inside maize (*Zea mays* L.). The proposed experiments will identify host genetic factors that affect response to endophytic organisms, link environmental factors to the communities of the leaf surface, and perform artificial selection to identify factors in the microbial community that are involved in a more positive outcome for the host plant (nitrogen acquisition).

Educational activities will foster student scientific identity through active learning with plant-associated microbes and mentoring students in scientific communication at a yearly professional conference. Public outreach will be carried out by bringing in national speakers to engage with the local community via a student-run outreach organization.

Intellectual merit

Plants and microbes have co-evolved for hundreds of millions of years, but the mechanistic details of their interactions are largely unknown. This project will advance our understanding of plant-microbe at multiple levels. It will analyze the effect of host plant genetics on interactions with endophytes (Aim 1), determine the environmental factors that shape microbes on the leaf surface (Aim 2), and analyze the effects of selection for improved plant-microbe interaction on the microbial community (Aim 3). These experiments cover the three major divisions of the plant microbiome (endopshere, phyllosphere, rhizosphere) and three of the largest forces shaping community makeup (host, environment, and selection). These experiments will identify the major drivers of microbial community assembly, quantify these drivers into mathematical models, and use these models to make testable predictions about community assembly and impact. In addition, these analyses will be highly comprehensive, spanning roughly 5000 plant genotypes (Aim 1) and at least 25 field locations across North America and the world (Aim 2), thus giving a broad insight into the mechanisms underlying plant-microbe interaction.

Broader impacts

Two projects will be undertaken to improve undergraduate retention in science-related careers. The first will focus on active learning and scientific investigation in a living laboratory. The second will involve mentoring undergraduates in both oral and written presentation skills at an academic research conference. National speakers will also be recruited to speak through a local student-run outreach organization, providing a rich scientific experience for the public and valuable leadership experience for the students involved.

One graduate student, a postdoc, and several undergrads will carry out the research in this proposal and will be trained in scientific research and presentation. They will gain expertise in plant genetic and bioinformatic approaches, learn to plan and implement independent research projects, be trained to effectively communicate their results, and gain their own experience as teachers and mentors. Emphasis will be on recruiting underrepresented groups, such as by recruiting undergrads through the NSF-funded Peach State Louis Stokes Alliance for Minority Participation (PS-LSAMP).

This research will be performed in maize, a highly important crop both in the United States and across the world. Understanding the fundamental mechanisms of its microbial associations could lead to more sustainable production practices and/or improved resilience in the face of climate change. This research will also create model systems for future study, define the core maize microbiome, and determine functional mechanisms behind specific plant-microbiome interactions, all of which are research priorities for harnessing plant microbiomes for human benefit[2]. As such, the insights from this project will likely be applicable to many other plant species, so that understanding the mechanisms of plant microbial assembly could have significant impact on agriculture, ecology, land restoration, biofuel production, or other fields involving plants in a natural or agricultural environment.

PROJECT SUMMARY

Instructions:
The summary is limited to 250 words. The names and affiliated organizations of all Project Directors/Principal Investigators (PD/PI) should be listed in addition to the title of the project. The summary should be a self-contained, specific description of the activity to be undertaken and should focus on: overall project goal(s) and supporting objectives; plans to accomplish project goal(s); and relevance of the project to the goals of the program. The importance of a concise, informative Project Summary cannot be overemphasized.

Title: Understanding the Disease Cycle of Exobasidium Leaf and Fruit Spot of Blueberry, and Development of Practical Disease Management Recommendations

PD: Harald Scherm	**Institution:** University of Georgia
CO-PD: William O. Cline	**Institution:** North Carolina State University
CO-PD: Phillip M. Brannen	**Institution:** University of Georgia
CO-PD:	**Institution:**
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The expansion of the blueberry industry has been an economic boon to rural areas across the U.S., but sustained growth is threatened by the emergence of new diseases. Exobasidium leaf and fruit spot, caused by the fungus Exobasidium maculosum, has become particularly damaging because its life cycle is unknown and control measures are lacking. This research-led project will collaborate with producers, the GA and NC blueberry commodity associations, the Southern Region Small Fruit Consortium (SRSFC), and Extension agents to better understand the life cycle of E. maculosum to develop practical disease controls. We hypothesize that the pathogen has a unique life cycle highly dependent on epiphytic survival of its yeast cells, and that this stage can be disrupted with simple disease management tactics that have added horticultural or pest management benefits.

We integrate three objectives, two focused on research and one on outreach:

1) Quantify survival of E. maculosum as epiphytic populations and in shoot lesions during summer, fall, and winter in naturally and artificially infected plantings;

2) Clarify how fruit become infected, viz. by basidiospores from leaf lesions or by overwintered yeast cells colonizing flower buds and flowers early in the season;

3) Develop and refine practical disease control recommendations and implement them in collaboration with Cooperative Extension and the SRSFC.

Program priority: CARE – "Develop and implement solutions to critical producer problems associated with … crop production, protection, or product quality."

Outcome: Robust and cost-effective control recommendations that will be implemented in collaboration with Cooperative Extension and the SRSFC.