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Analysis of Horizontal Gene Transfer Mechanisms in Soil Microbiomes

Tatyana Sysoeva

RCEU21-BYS-TS-02

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Project Title - Analysis of horizontal gene transfer mechanisms in soil microbiomes

Faculty - Tatyana (Tanya) Sysoeva, Assistant Professor, Department of Biological Sciences, Shelby Center room 369M, University of Alabama in Huntsville, 256-824-6371, tatyana.sysoeva@uah.edu

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Project Description - Antibiotic resistance is an increasing global health threat that is aggravated by apparent stagnation in process of new antibiotics development. Horizontal gene transfer is a main contributor to the spread of antibiotic resistance genes amongst bacteria, and proceeds through conjugation, transduction or natural transformation. Conjugation requires two cells to interact to transfer resistance genes, while natural transformation allows bacteria take up extracellular naked DNA. While the processes of conjugation and transformation are well understood in some human pathogens, it is unclear how these processes happen in more complex systems such as mixed microbiomes. In this project on analysis of horizontal gene transfer in soil microbiome we will focus on identifying novel transformable species and classes of conjugative plasmids in soil mixed communities. Moreover, such complex microbiomes often exist as surface biofilms – multilayer communities of microbes on solid surfaces. It is unclear how the biofilm environment would affect the gene transfer. To analyze present transformable species and conjugative elements we will a combination of microbiological, molecular and next generation sequencing methods. In result we will establish the diversity of species and elements capable of horizontal gene transfer and potentially contributing to the spread of antibiotic resistance genes in environment.

Student Duties, Tangible Contributions, and Specific Outcomes - This project will require full time presence in the lab for 32-40 hours per week, for 10-12 weeks. During this time the student

will learn basic microbiological procedures and several functional assays. In particular, this project will entail preparing solutions and plates; working with uropathogenic *Escherichia coli*; grow biofilms; conduct conjugation assays; and image bacteria using confocal microscope. In addition to practical work, the researcher will read the peer-reviewed scientific articles that is required for understanding and developing of the project at hand. The student will also learn how to document the experimental work and keep laboratory notebook with the detailed description of the performed experiments. The student will be encouraged to prepare a poster for consecutive presentation at a scientific conference. At the end, the student researcher will be required to write a report with suggestions for future development of the project and analyses of the obtained results that can be used in a future publication. This report will be presented at a lab meeting to share the results and experiences with the whole group.

Student Selection Criteria - The student (from freshman to senior) should have completed some basic biology courses at UAH or as AP courses. While a good academic standing is not a requirement, the applicant has to have a sincere interest in the topic of microbiology and willingness to learn molecular techniques.

Project Mentorship - Dr. Sysoeva will provide one-on-one meetings to introduce to the project, supervise literature review, discussions and analyses of the experimental results. In addition, weekly group meetings will be held to share the results and solve any ongoing issues. She will ensure close daily supervision and training in the laboratory setting for the hands-on work at the bench, including safety training and microbiological work. Dr. Sysoeva will guide the student researcher in writing up the report of the obtained results and preparing for the final presentation.

In addition, four MS students and an experienced undergraduate researcher working on related projects will be conducting their research in the laboratory and assist with microbiological work, such as media preparation, autoclaving, inoculations. Two of the MS students have experience teaching microbiology. It is particularly important that one of the MS students has recently completed the summer RCEU project and thus is familiar with the program goals and requirements.

Safety and Contingency Plan - Student awarded will complete required general lab safety and biosafety trainings at the UAH OEHS website. This would include any COVID-19 related training as well. As part of our research group the RCEU student will follow the developed reduced density standard procedures that were approved by the COS earlier in 2020 to allow the re-start of the wet lab work in the summer of 2020.

If due to public health concerns the experimental wet lab work on the UAH campus would not be allowed, the next RCEU project will take place. We will proceed with bioinformatic analysis of the DNA uptake genes in several bacterial genomes, e.g. lactobacilli species which we recently sequenced. These analyses of DNA uptake operons *in silico* will identify potential regulators of the DNA uptake process and future route of testing for this process in the lab. The outcome of this bioinformatic project will be a write-up of the DNA uptake operon analysis and proposed plan of testing for active DNA transformation in the analyzed species.