

November 14, 2022

Undergraduate Research Initiative Committee
University of Rhode Island
Kingston, RI 02881

Dear Committee Members,

I am extremely pleased to offer my support to Aisling Macaraeg for her undergraduate research project, "Discovery of Genes Involved in *F. tularensis* Survival in Freshwater." My laboratory studies the molecular mechanisms that lead to pathogenicity of the human pathogen *Francisella tularensis*. While this organism is highly pathogenic to humans, my laboratory takes advantage of the model organism *F. tularensis* subsp. *holarctica* LVS (Live Vaccine Strain), which does not infect or cause disease in humans and we do not work with any of the highly pathogenic strains.

Aisling is an academically outstanding upper-level student, with GPA of 3.91, and has been an active researcher in my laboratory since the fall of 2021. On her first research project, she worked collaboratively with another undergraduate researcher, a project ultimately funded by a (URI)² undergraduate research grant. That work focused on understanding the regulation of a ribosomal protein gene. The results that Aisling and her collaborator obtained revealed unexpected complexity that initiated a new line of investigation in my laboratory currently pursued by a Master's student.

In the spring of 2022, Aisling worked both on the ribosomal gene regulation project as well as her independent honors project. The goal of her independent honors project was to identify what laboratory conditions might mimic the long-term persistence of *F. tularensis* in freshwater in the environment. We know very little about the survival of *F. tularensis* in the environment, so this project provided critical initial data to create a lab-based model for *F. tularensis* environmental persistence. Aisling successfully completed multiple months-long experiments to obtain these data during the spring semester and over the summer, during her RI-INBRE Summer Undergraduate Research Fellowship (SURF).

Aisling is interested in further exploring which specific genes allow *F. tularensis* to persist over long periods of time (21 – 56 days) in freshwater. In our discussions about this project, Aisling has demonstrated that she is aware of prior research identifying a specific gene necessary for *F. tularensis* to survive the transition from high to low osmolarity, as occurs when *F. tularensis* enters the freshwater environment. She hypothesized that additional genes may be necessary, a hypothesis we can test using a state-of-the-art approach referred to as "Tn-Seq." Aisling's work over the summer has already prepared her to use this approach, as she modified a genetic tool to make downstream steps more cost-efficient. I am an expert in the use of the Tn-Seq method, so am well-qualified to train Aisling. I enthusiastically support the continued development and undertaking of her independent research project.

Together with my graduate students, I will supervise all the proposed work in this project to ensure safety, rigor, and reproducibility. The success of this project will be based on Aisling's ability to (i) learn and implement research protocols, (ii) critically analyze the resulting data, (iii)

make connections between the resulting data and the current scientific literature, and (iv) effectively communicate research findings, both within and outside our research group.

Aisling is an outstanding student with great enthusiasm for laboratory research. Her undergraduate research experience in my laboratory, including the exciting work proposed here, will prepare her to achieve her future goal of entering graduate school in biomedical research. She is extremely well-prepared to carry out the proposed experiments and I support her application for an undergraduate grant for original student research without reservation and with enthusiasm.

Sincerely,

A handwritten signature in dark ink, appearing to be 'KR' followed by a horizontal flourish.

Kathryn M. Ramsey, PhD
University of Rhode Island
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