Budget Justification

Senior Personnel

The University of Idaho has determined the salary year for senior personnel to be based on the calendar year. A 3% increase in senior personnel salaries has been calculated in years 2-5 to account for anticipated inflation.

Paul Rowley, Principal Investigator, 1 calendar month per year

An Assistant Professor of Biological Sciences at the University of Idaho. Dr. Rowley will oversee overall project direction and will mentor and train the graduate student and technician in techniques that are relevant to microbiology, molecular cloning, and genetics. Dr. Rowley will also support the career development of personnel by providing opportunities for scientific communication, outreach, and additional skills training through courses and professional workshops at the University of Idaho. Dr. Rowley will also be responsible for training all personnel in safe laboratory practices and techniques pertinent to biological safety level 2. Dr. Rowley will be primarily responsible for writing project progress reports and manuscripts that result from data collected during the project. Requested amount is $44,034 total.

Other Personnel

Lance Fredericks, Technician, 12 calendar months per year

As the current senior laboratory technician in the Rowley laboratory he has been an essential member of the killer yeast research team for the last two years. Mr. Fredericks has extensive experience working with killer yeasts and will directly support the empirical research efforts of the graduate and undergraduate students in the laboratory. Initially this will involve training the students in specialized techniques that he has mastered in the Rowley laboratory for the handling and maintenance of killer yeasts and the purification of killer toxins in year 1. In the following years he will focus on the completion of **Objective 2** and will provide additional help and advice to the students that are working on **Objectives 1 and 3.** Mr. Fredericks is experienced in team management and student mentoring having previously directed teams of 4-5 undergraduate on projects that have led to the collection of all the preliminary data for this proposal. A 3% increase in staff salaries has been calculated in years 2-5 to account for anticipated inflation. Requested amount is $191,128 total.

Jack Creagh, Research Assistant, 12 calendar months per year

A current 2nd year graduate student in the Rowley laboratory who has worked previously on projects involving the identification and characterization of extracellular virus particles in *Saccharomyces* yeasts. His efforts will be focused on the completion of the experiments of **Objective 1**. This will provide sufficient opportunity to collect data for publication and completion off his thesis and graduation in year 4 or 5 of the project. He is a good fit for this objective because has a strong prior research experience working with yeasts, excellent aseptic technique, and skills in molecular biology. He will also work collaboratively with Mr. Fredericks and Ms. Garcia to achieve broader project goals when needed. Mr. Creagh will be directly mentored by Dr. Rowley to ensure the completion of project goals and adequate progress towards his PhD thesis. We request 12 calendar months of salary per year. Requested amount is $125,000 total.

Ximena Garcia, Undergraduate Student, 12 calendar months per year. A junior undergraduate researcher in the Rowley laboratory who is skilled in microbiology and the discovery of killer yeasts. She has worked in the laboratory for 8 months and has made several interesting discoveries regarding the application of killer yeasts against spoilage organisms and

has discovered a new type of killer toxin. As a female underrepresented minority (Latino heritage), I believe that she is well placed to lead a cohort of undergraduates in killer yeast discovery as described in **Objective 3** and our **Outreach Activities** to will serve as a role model for other underrepresented students recruited from the TRIO and McNair programs at the University of Idaho. Her responsibilities will include training undergraduates in microbiology techniques and ensuring that experiments are performed rigorously and safely. She will also be responsible for the overall management of the culture collection of novel killer yeasts. Requested amount is $33,800 total.

Fringe Benefits
University of Idaho FY22 consolidated fringe rates applied. 29.4% for faculty, 40.8% for staff and 3% for students. Total fringe benefits requested for this project is $95,690.

Travel
We request a total of $4,482 for domestic travel.

Data generated by this project will have broad significance to the field of microbiology. For this reason, we plan to attend the yearly conference ASM Microbe, which is held in the US. The conference is attended by thousands of scientists every year and includes sessions relevant to many aspects of this project, such as Applied and Environmental Science, Ecological and Evolutionary Science, Host-Microbe Biology and Molecular Biology and Physiology. We anticipate that we will present results from this project in year 2 and 4. The estimated costs are based on attending the conference in 2024 in Atlanta (Georgia) and include registration fee, domestic airfare and accommodation for the 4 days of conference activities and two days travelling. Per diem is calculated based on U.S. General Services Administration per diem rates (Budget table 1).

	Year 2	Year 4
Accommodation ($240/day)	960	960
per diem ($61/day)	366	366
Economy airfare	390	390
Conference registration	525	525
Annual total ($)	2,241	2,241
	Grand total ($)	**4,482**

Budget Table 1.

Breakdown of estimated costs associated with attending ASM Microbe conference in 2024 and 2026.

Other Direct Costs

Materials and Supplies

Total Materials and Supplies requested amount is $41,877.

$8,000 per year is requested for lab supplies. All work performed in the Rowley lab is rooted in the fundamental techniques of microbiology, thus requiring adequate supplies for the culturing of microorganisms, including agar plates, media, buffers, disposable pins for robotics, and sterile plasticware. In addition, we require reagents for molecular biology, including restriction enzymes, polymerases, ligases, TOPO-TA cloning vectors, agarose powder (electrophoresis grade) and molecular biology grade chemicals and buffers. The average cost of project reagents accurately reflects the nature of the work proposed is based on the average spend rate of currently administered projects within the Rowley laboratory.

$1,242 is requested in year 2 for Gateway cloning reagents sufficient to clone all 65 mutant oligo pools constructed by Twist biosciences into a stable plasmid vector for the creation of PCR cassettes for yeast mutagenesis and long-term storage.

$635 is requested in year 1 for a portable trinocular biological compound microscope with plan objectives + 5MP USB3.0 Digital Camera that will be transported to middle and high schools' classrooms that lack microscopes for the visualization of yeast cells that students isolate from the environment.

Publication Fees
$1,000 in years 3-5 is requested to defray the cost of publishing the project's research.

Consultant Services

Total Consultant Services requested amount is $11,650.

$2,100 is requested in years 2-5 for Illumina Sequencing of amplicon sequencing libraries that will be used to monitor the prevalence of a pool of 1,235 Kre1p mutants in mixed culture. Sequencing funds will also support the discovery of novel viruses and satellites discovered by undergraduate researchers as part of the educational objectives (MiSeq v2 kit (Nano 500 cycles) ~ 1 million reads).

$3,250 is requested in year 2 for a site-saturated mutagenic library of Kre1p obtained from Twist biosciences.

Other

$500 per year is requested for outreach and educational supplies that will include agar plates, microscope supplies, general media for yeast growth and enrichment. These supplies will be sufficient to run outreach activities over all three outreach locations in Moscow Idaho.

We also include $100 per year for purchase of plush Giant Microbe™ scale models of the yeast *S. cerevisiae.* These will be used as incentives for survey participants at the Moscow farmer's market over 6 weeks (6 per week).

$68,114 total is requested for the project research assistant's tuition/fees and insurance. A 5% anticipated annual inflation increase has been applied in years 2-5.

Indirect Costs
Calculated at the University of Idaho Federally Negotiated Indirect Cost Rate as Modified Total Direct Cost excluding participant support costs, equipment > $5,000, tuition, and those amounts of sub awards in excess of $25,000. 48.5% rate is applied to the first three months of year 1. 50% rate is applied to the remaining 9 months of year 1 and all of years 2-5. Total F&A for this project is requested in the amount of $276,445.