Microbial population at Washington State University Vancouver campus: A course-based undergraduate research experience in microbiology

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Research Objectives

- To examine if bringing an independent research project into the required coursework enhances student learning experience
- To characterize and identify bacterial populations present at different buildings at WSU Vancouver

Introduction

Undergraduate research experiences (UREs) have shown to increase students' ability to engage in scientific practices such as learning to think like a scientist, provide leverage towards pursuing graduate education or careers in science,[2,3] and is positively correlated with higher education graduation rates.[4] However, most of the undergraduate students do not get an opportunity to participate in scientific research; which is an essential component of undergraduate training in biology.[5]

Course-based undergraduate research experience (CURE) provides research opportunities to more students, which are inherently limited, on campus. In addition, due to research being a part of their required lab coursework, more students are involved despite any personal or financial burdens.

The purpose of this pilot study was to assess the impact of a microbiology CURE on student understanding of what it means to 'think like a scientist' and learning gains. We also hoped to identify bacterial populations at heavily used buildings on our campus. Student response to what it means to 'think like a scientist' and how student 'thinking like a scientist' changed after the research project, were categorized. The skill gains through CURE participation were measured by student self-identification of learning items described in the Survey of Undergraduate Research Experiences (SURE)[6] and the related CURE survey. To the best of our knowledge, this is the first-time bacterial population on our campus buildings was investigated.

Methods

Participants and Sample collection. The study was conducted with Microbiology laboratory (MBioS 306) students, spring 2019, at WSU Vancouver. A survey was conducted in which students identified the most heavily used buildings/areas on WSU Vancouver campus. Students collected samples. At the end of the course, students presented the comparison of their sample results to the identified genus using morphological and biochemical characteristics. n= 21. One lab section.

Microbiology CURE Survey. A post-course CURE survey was administered. The CURE survey results includes student understanding of 'thinking like a scientist', how student 'thinking like a scientist' changed after taking this course, and learning gains from the research project. The CURE survey was adapted from the previous studies.[5,6] The SURE survey[6] data represents 1,135 students who completed summer and fall research in 2003. Likert scale data was plotted as a diverging stacked bar and scatter plot showing medians with interquartile range (IQR) error bars.

Staining and Biochemical Testing. All stains were performed using control organisms on the same slide. Phenylethyl alcohol agar (PEA), Mannitol Salt Agar (MSA), MacConkey (MAC) agar, Eosin Methylene Blue (EMB) agar, and Bile Esculin Agar (BEA) were performed. Differential media tests such as citrate utilization test, Kligler Iron Agar (KIA), phenol red broth, and tests detecting enzymes such as catalase, oxidase, amylase, and urease tests were performed. Aerotolerance was determined using Fluid Thioglycollate (FTG) and Oxidative-Fermentative (O-F) tests. Controls were used.

DNA Extraction, PCR Amplification, and Sequencing. DNA was extracted using Qiagen DNeasy blood/tissue kit followed by PCR amplification of universal bacterial 16S rRNA gene. DNA concentration and purity was determined using a Nanodrop. Agarose gel electrophoresis was performed. PCR samples were sent to Vollum Institute at Oregon Health and Science University (OHSU) for Sanger sequencing. Eleven student samples remained unidentified during the semester. The unidentified samples were later processed and identified by 5 undergraduate students from the spring 2019 course.

Safety. Students followed all the necessary safety procedures. This included no open-toed shoes, no food/drinks, use of gloves and bunsen burners, informing and cleaning any spills immediately with a disinfectant, and exposure to UV light for any spore-forming bacteria.

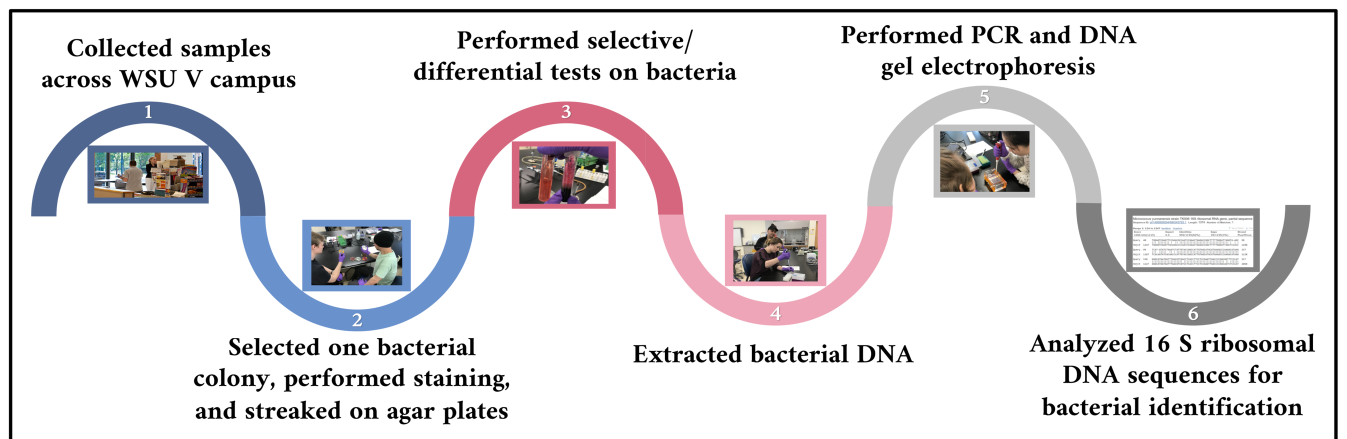

Figure 1. Workflow and research methods in the MBioS 306 Microbiology Research Project.

Figure 2. MBioS 306 Microbiology laboratory students contributing to the Research Project. n = 21

Results

Table 1. Student response to what it means to 'think like a scientist' and how student 'thinking like scientist' changed after the research project.

Category	Definition	Percentage response to 'what it means to think like a scientist'	Percentage response to 'how thinking like a scientist changed after conducting research project'
Questioning	Use of the word; challenging all the possibilities	43.75	11.76
Follow scientific method	Describing a part and/or process of scientific investigation such as following protocols, performing experiment, data analysis; future steps depend on previous results; use of the word	43.75	5.88
Open-mindedness	Use of the word; understand that failed results occur; don't assume results; not being upset with non-significant results	25	23.53
Critical thinking	Use of the word; trying to find why data failed; troubleshooting and problem-solving	31.25	17.64
Scientific communication	Communication of the results	6.25	NI
Understanding scientific process	Increased knowledge and understanding of science and research work; failure occurs; dig deeper for information	NI	47.05
Microbiology comprehension	General awareness of surroundings; microbes around us; survival of microbes; characteristics of bacteria	NI	29.41

NI = Not identified. Categories were first identified individually and later discussed and defined collectively by three people.

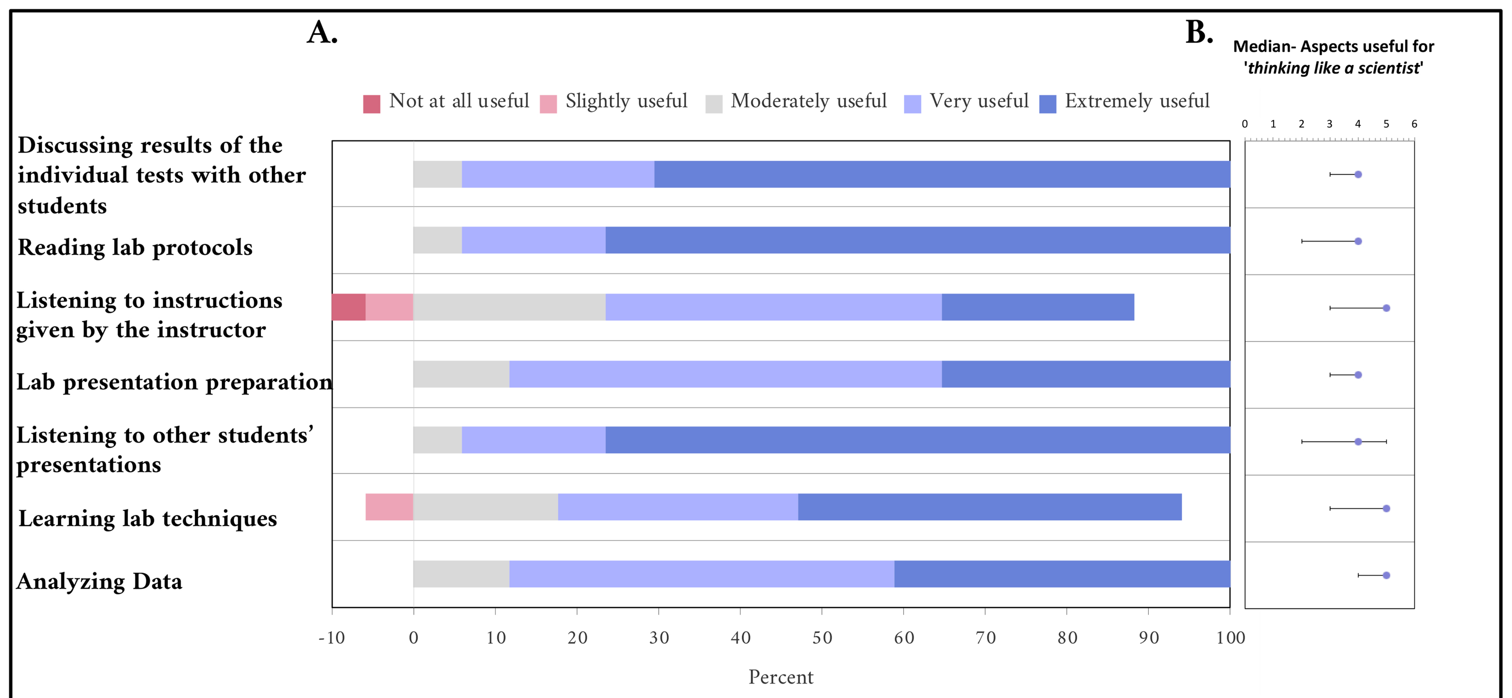

Figure 3. Aspects of the research project that were most useful for improving your "thinking like a scientist." A total of 7 learning items were evaluated by the students as '5- extremely useful, 4- very useful, 3- moderately useful, 2- slightly useful, and 1- not at all useful' on a closed-ended Likert scale. **A.** Diverging stacked bar showing the percentage of the responses to each item. **B.** Scatter plot showing the medians for each learning item[5] for MBioS 306 Research Project (purple circles). IQR for error bars. n = 17.

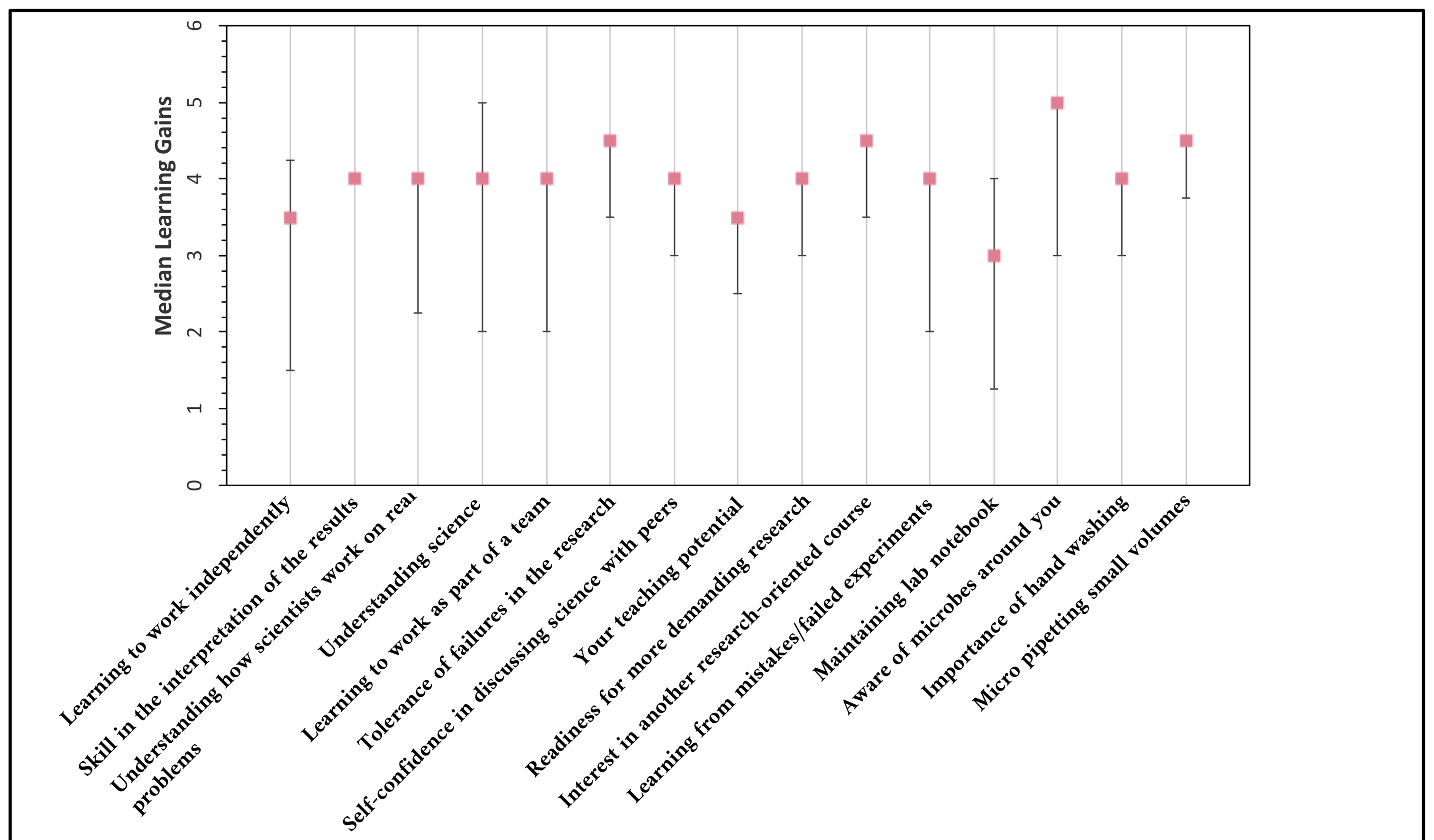

Figure 4. Student evaluation of learning gains. A total of 15 items were evaluated by 17 students following the spring 2019 semester completion as '5-very large gain, 4-large gain, 3-moderate gain, 2-small gain, and 1-no gain or very small gain' on a closed-ended Likert scale. Scatter plot showing the median learning gains for each item MBios 306 Research Project (red squares). Learning gain items were taken from SURE[6] data. The last five items were selected based on course relevance. IQR error bars are shown.

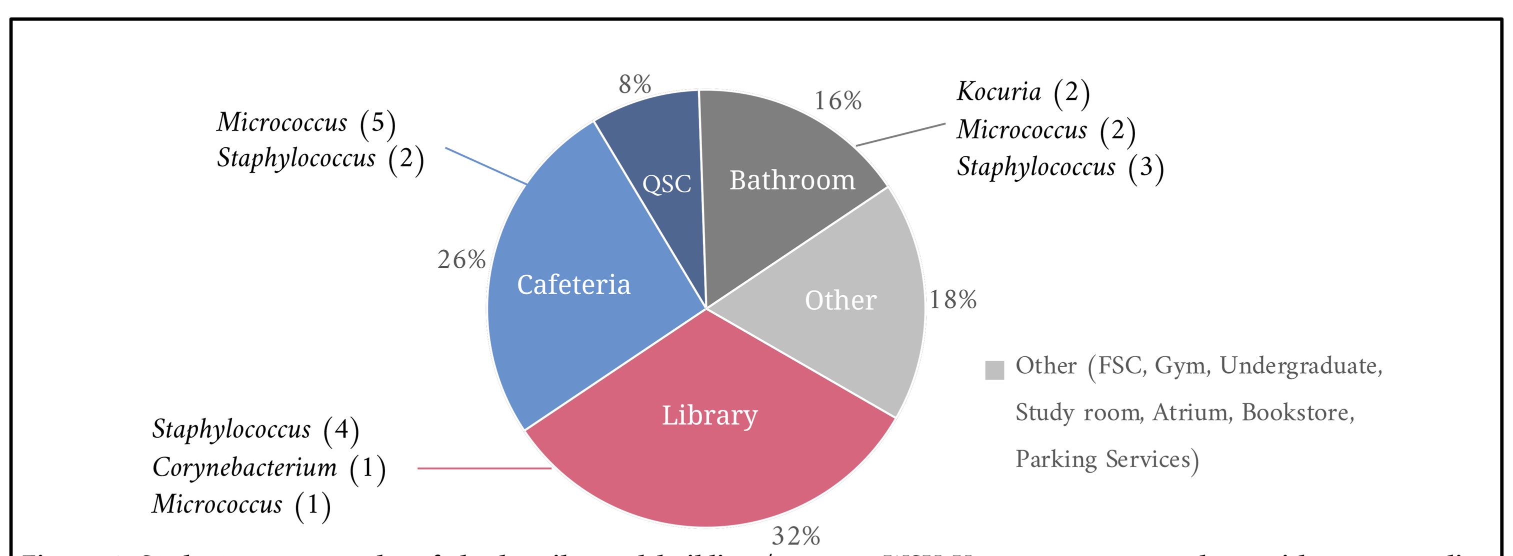

Figure 5. Student survey results of the heavily used buildings/areas on WSU Vancouver campus along with corresponding bacterial genus identified at the sample collection site. The most heavily used buildings/areas identified by the students were used for sampling. The percentages depict the number of student responses for the building/area. The sampling areas chosen at the buildings were places that are commonly touched such as computer keyboards, vending machines, and bathroom doors. Each student chose a different sampling area. FSC = Firstenburg Student Commons, QSC = Quantitative Skills Center, Atrium = Student area in the Sciences building. Genus' results are mentioned corresponding to its location. The number next to each genus indicates the number of samples resulting in the same genus identification from different areas at the same building. One student sample remained unidentified. n = 21.

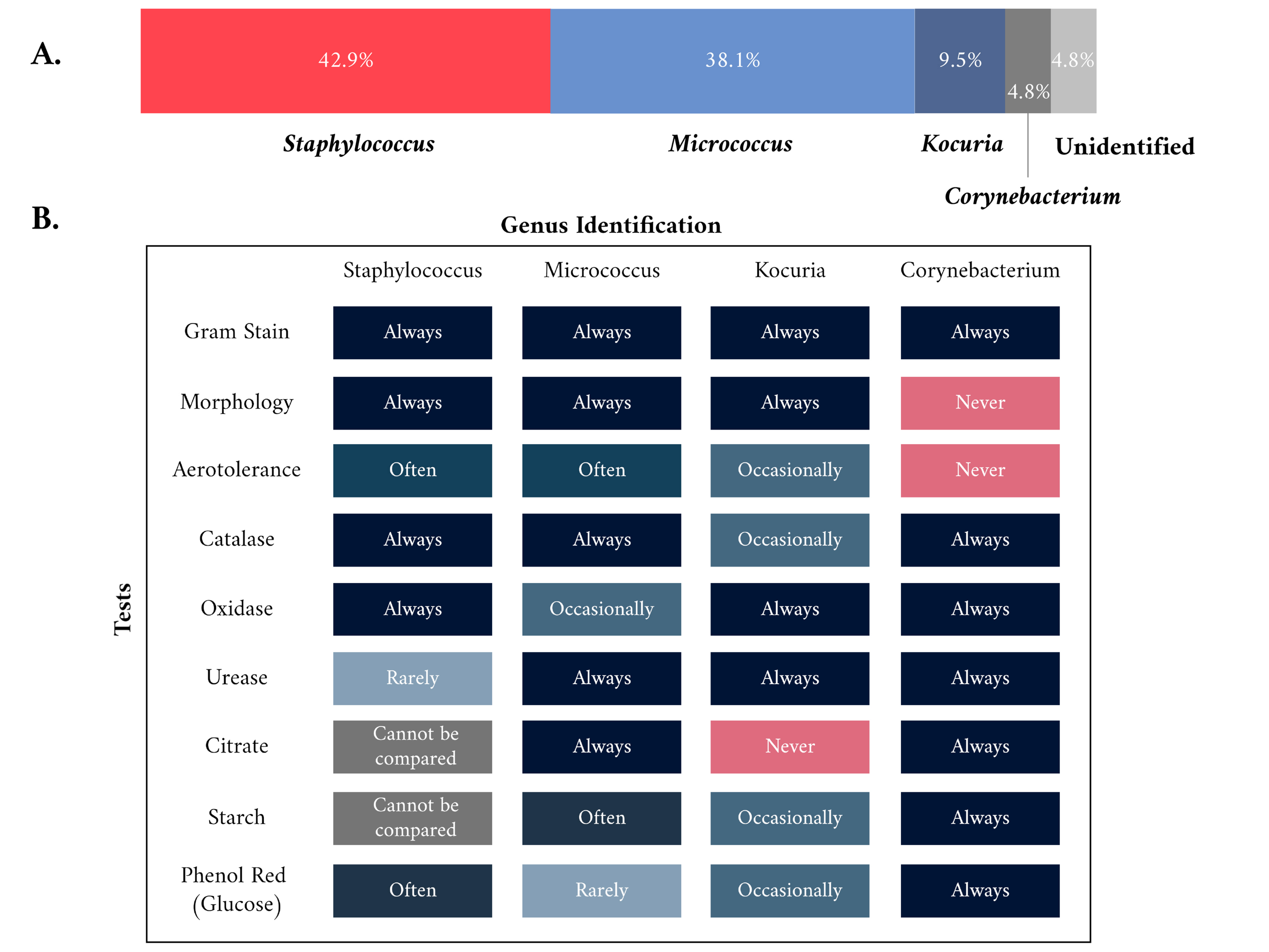

Figure 6. Percentages of genus identification, and association of biochemical tests of student samples to the identified genus. A. Percentages depicting student samples identified as *Staphylococcus*, *Micrococcus*, *Kocuria*, and *Corynebacterium* and one unidentified sample based on 16 S ribosomal gene sequencing. **B.** Students' test results compared to known genus characteristics were categorized based on percentage of samples matching expected results (always = 100%, often = 66-99.9%, occasionally = 33-65.9%, rarely = 0.1-32.9%, and never = 0%). "Cannot be compared" refers to characteristics that are highly variable within the indicated genus and thus are unable to be compared. Other tests as mentioned in the methods section were performed but were not found useful for associations. n = 9, 8, 2, and 1 for *Staphylococcus*, *Micrococcus*, *Kocuria*, and *Corynebacterium*, respectively.

Analysis and Conclusion

This pilot course-based undergraduate research experience allowed 21 undergraduate students to participate in microbiology research. In an open-ended post-course survey, students mentioned questioning, following scientific method, and critical thinking are important components of thinking like a scientist. Students reported that understanding of scientific process, open-mindedness, and microbiology comprehension were important when thinking like a scientist during the research project. After the course, students showed a large gain in tolerance to failures in research and awareness of microbes around them. Students also exhibited an interest in taking another research-oriented course.

In 95.2% cases, 16S rRNA provided genus identification; however, variability was seen at the species level. Sanger sequencing from different student samples identified *Staphylococcus, Micrococcus, Kocuria,* and *Corynebacterium* genus; all predominantly present on human skin. The comparison of actual biochemical test results to the expected biochemical test results based on the genus identification by sequencing yielded in delineating reliable biochemical tests. For example, catalase test was reliable for the identification of *Staphylococcus*, *Micrococcus*, and *Corynebacterium* with the student results matching the expected results 100% of the time. In contrast, tests such as phenol red with glucose were unreliable when comparing expected to actual results for *Micrococcus* (25%) and *Kocuria* (50%).

The lab presentations given by students involved comparison of actual and expected biochemical tests based on genus identification was challenging, and required critical thinking and literature search. We show that CURE implementation into microbiology coursework yields students with better research skills, generates scientific data, and a high level of interest towards their surroundings and microbes. The gains from this pilot CURE suggest that it should be implemented in more undergraduate laboratory courses.

Lessons Learned and Future Directions

- Optimization of the protocols before the course starts. This will avoid loss of time, energy, and resources during the course.
- Videos demonstrating micropipette techniques and/or individual lab demonstrations will ensure fewer sampling errors.
- Both pre- and post-course surveys will allow comparisons of the learning gains.
- Future directions would include the above lessons learned and tests that can reliably help in differentiating gram-positive organisms such as fermentation of different sugars, coagulase test, and antibiotic susceptibility testing.

Acknowledgements

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