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Identification of Bacteriophages that Target *Staphylococcus aureus* MRSA strains

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Staphylococcus aureus is an opportunistic pathogen that is common in hospital and community settings. These bacteria are dangerous because they can cause illnesses such as sepsis, meningitis, and pneumonia, while also being difficult to treat due to living inside biofilms, which are communities of bacteria that live on surfaces, and quickly adapting which leads to antibiotic resistance [1]. Together, these two traits make conventional antibiotic treatments less or completely ineffective. The most clinically significant strain of *S. aureus* is Methicillin Resistant *Staphylococcus aureus* (MRSA). MRSA is an antibiotic resistant strain that is notoriously difficult to treat and is the leading cause of bacteremia in the world [1]. Its prevalence and danger push research into develop alternative therapies that do not include antibiotics. One promising area of research is the utilization of bacteriophages (phages). Phages are viruses that have evolutionally evolved to target bacteria [2]. In lab settings, phages have demonstrated the ability to target resistant strains of bacteria such as MRSA, while also being abundant in the environment and safe to humans [2].

The purpose of my research was to investigate if phages with the ability to target MRSA are present in the local environment. Through this process, my specific aims were to: 1) isolate bacteriophages from the environment with specificity against MRSA, 2) determine the effectiveness of isolated phages against multiple strains, or variations, of MRSA, and 3) investigate if the isolated phages have synergy and are more effective at treating MRSA when combined with the antibiotic vancomycin.

Before sample collection, one naturally occurring wild type *S. aureus* strain and a panel of 7 clinical MRSA strains isolated from patients in hospital settings were grown in liquid culture. In addition, various reagents such as a phage isolation buffer were made following a protocol from similar research found during a literature review [3]. Environmental samples being used to find phages were collected from two sources. The first source is the Branford Trolley Trail in Branford, Connecticut where water and mud samples were obtained from the waterways surrounding the path. The second source of samples were untreated sewage samples collected from the Meriden Sewage Treatment Plant and the Branford Water Pollution Control Facility.

After collection, each sample was tested to determine the presence of phages that have the ability to target one wild type and one resistant strain of *S. aureus*. In order to test these samples, a procedure known as the top layer agar method was performed [3]. During this protocol, each sample was combined with the phage buffer before being mixed with an aliquot of *S. aureus* culture and molten agar, and then poured onto a standard Tryptic Soy Agar (TSA) plate. This setup provided the nutrients required for the bacteria to grow and enabled the observation of any bacteriophages present.

Final results demonstrated that bacteriophages were present in one of the sewage samples collected from the Branford Water Pollution Control Facility. This discovery showed that viruses with the ability to specifically target one MRSA strain could be successfully obtained from the environment. Isolating this virus presents an opportunity to develop a therapy that can be used to target resistant strains of *S. aureus*. Future research will involve testing its effectiveness against

the other MRSA strains in the clinical panel, its synergy with antibiotics, followed by sequencing to determine its genetic make-up.

References

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