Te Niwha

Research Project Impact Case Study

Development of non-invasive "liquid-biopsy" methods for Infectious disease using microbial cell-free DNA

Short Research Title

Creating simple blood and urine tests to detect infections using microbial DNA

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Introduction

Infectious diseases are traditionally diagnosed by culturing the microorganism causing disease. However, in many cases a diagnosis can be difficult to achieve because of inability to obtain the appropriate invasive specimen, non-viability of the micro-organism (for example due to antibiotic pre-treatment) or inability to reliably grow the organism in a laboratory environment. When micro-organisms cause disease in the body they release bits of their own DNA, and this can be detected in the blood and urine of patients. Our project set out to determine if two assays (one for blood and one for urine) can detect the microorganism DNA released during infection by quantitative polymerase chain reaction (qPCR). The two assays rely on non-invasive samples that are easy to obtain and would allow for quick infection diagnosis and treatment. Using either of these tests in the clinical setting will benefit all New Zealander's including our Māori and Pasifika communities.

We are investigating two different methods to detect microbial DNA in patient blood and urine. One approach uses magnetic beads coated with special DNA probes to capture pathogen DNA for detection by qPCR. The other uses CRISPR technology to detect microbial DNA in patient samples after qPCR amplification. The initial application of both assays has been for the detection of *Legionella* bacteria which causes a type of pneumonia called Legionnaires' disease. While the initial work is on the improved detection of Legionnaires' disease these two assays can be modified to detect any infectious disease as long as the microorganism's sequence is known.

This work has been made possible by collaborations with Canterbury Health Laboratories, Canterbury Respiratory Research Group and the Te Whatu Ora Waitaha Canterbury Paediatrics Department. These collaborations align with the Te Niwha Investment Objectives by strengthening collaborations between researchers, clinicians and health agencies while building on New Zealand's capability to demonstrate research excellence for infectious disease research and research translation. Since both platforms can be used for any infectious disease detection this project contributes to increasing New Zealand's preparedness and readiness for future infectious disease outbreaks. Likewise, the CRISPR assay has the potential to be developed into a point of care system that can contribute to improving health outcomes and increasing equality in low resource settings.

Results

Our two tests are highly sensitive and can detect *Legionella* down to low levels. The hybridization assay qPCR can detect as little as 10 copies of bacterial DNA (Figure 1a), while the CRISPR-based test can identify just one bacterial genome per test (Figure 1b). Both tests have shown to be highly accurate in the lab, correctly identifying only *Legionella* bacteria while testing against more than 30 other types of bacteria.

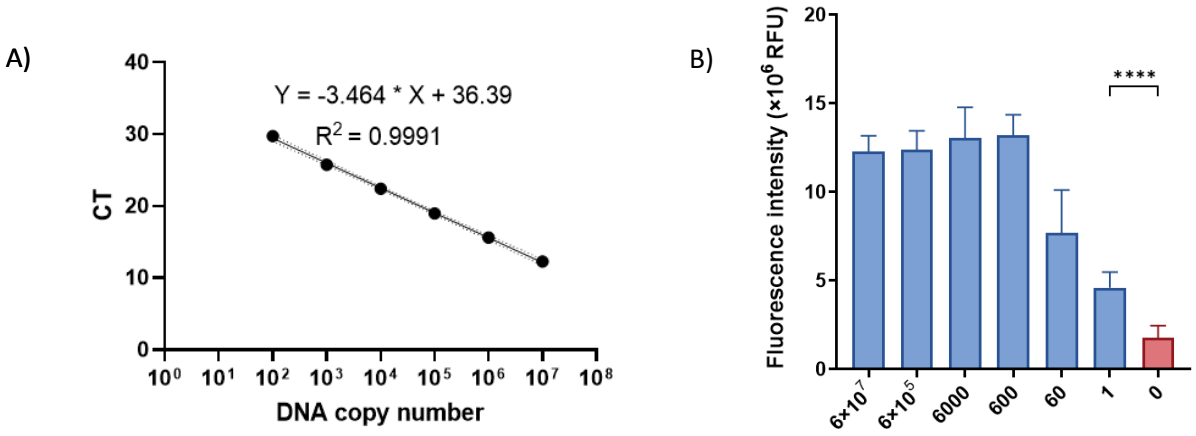

Figure 1. A) Limit of detection for hybridisation qPCR and B) limit of detection of CRISPR assay.

Our next step is to test real patient samples to determine their effectiveness in diagnosing true *Legionella* infections in a group of patients with pneumonia. We have collected over 500 blood and urine samples from patients admitted to Te Whatu Ora Waitaha Canterbury with pneumonia over three *Legionella* seasons which will be tested. The results will then be compared to the clinical and microbiological diagnosis to determine the accuracy of our two tests.

Impact

Early detection of infectious diseases using non-invasive methods and accurately determining the true burden of infection can significantly reduce healthcare costs, improve patient outcomes, and enhance public health, particularly for disadvantaged groups. By identifying infections early, costly hospitalizations and invasive procedures can be minimized, while timely treatment prevents complications and limits disease transmission. Accurate burden estimation supports better resource allocation and disease surveillance, reducing antimicrobial resistance and improving long-term management. Non-invasive tests also enhance accessibility, particularly for underserved populations, such as Māori and Pasifika, helping to close health equity gaps and ensure earlier diagnosis and treatment. These benefits collectively lead to lower healthcare costs, improved population health, and better health outcomes for high-risk and disadvantaged communities.

The ease of sample collection means these two tests have the potential to improve early and accurate diagnosis of many infectious diseases, leading to better treatment and outcomes for patients, while reducing cost on the public healthcare system.

The next phase of this work will involve testing all clinical samples using both platforms which will allow for the comparison between the two platforms and the clinical diagnosis. Future direction for the CRISPR-Cas assay is to remove the qPCR step by using circular DNA lowing equipment costs, simplifying workflows and reducing the need for experienced laboratory staff. This would then allow for the trial of this assay as a point-of-care test in rural and remote communities such as the far north.