Development of a point-of-care tool for diagnosing coral reef health

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revive & restore
GENETIC RESCUE OF ENDANGERED AND EXTINCT SPECIES

Abstract

- Mass coral die-offs globally from disease, climate change, and external stressors
- No current tools available for rapid and early detection of stress or disease
- Nucleic acid-based detection methods are reliable and easily adjustable for new targets
- Current methods require expensive equipment and reagents, making use more difficult in low resource areas
- Developed a portable point-of-care tool pipeline for loop-mediated isothermal amplification (LAMP) and electrical impedance-based detection

Background

- As many as 1 billion people rely on coral reefs globally for the services their ecosystems provide
- Anthropogenic consequences are weakening their health and complicating conservation efforts
- Stress can cause the release of the coral endosymbiont (bleaching)
- Disease outbreaks have caused massive population declines
- Climate change likely has impacted mass spawning timing and caused asynchrony, limiting repopulation

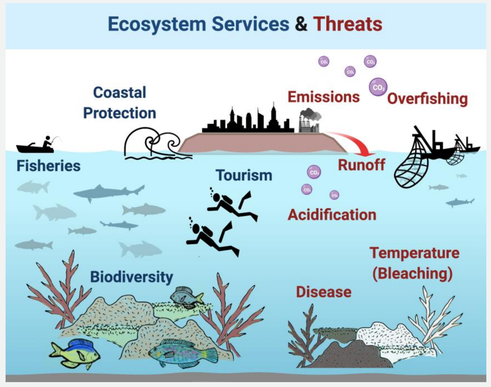

Figure 1. Services from and threats to coral reefs currently.

Methods

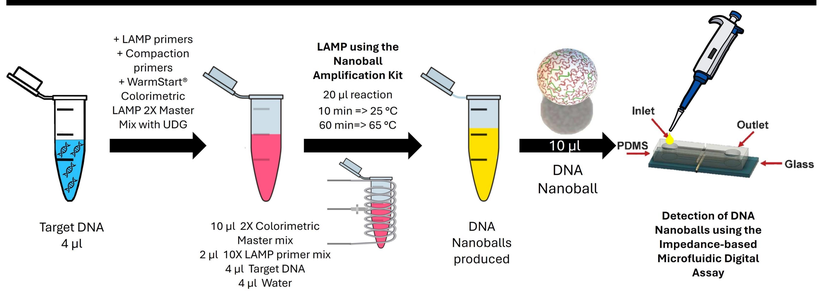

Figure 2. Protocol of the DNA nanoball LAMP amplification kit for a positive sample reaction.

Results

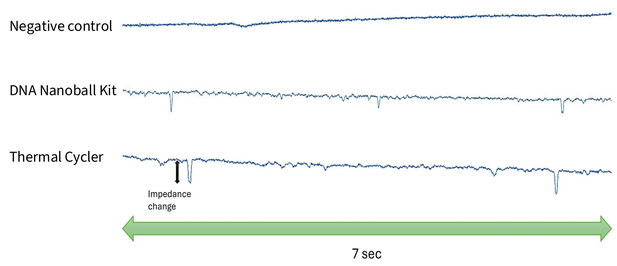

Figure 3. Electrical impedance readings for amplified samples.

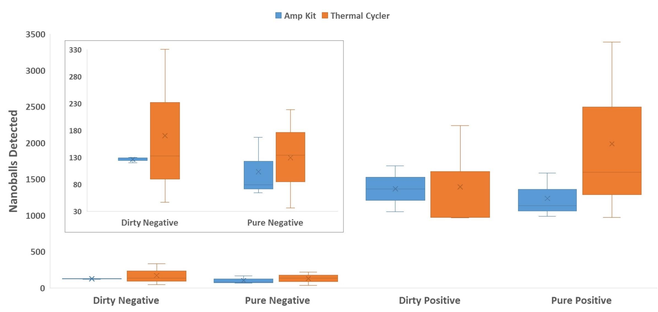

Figure 4. Number of nanoballs produced from amplified samples. Dirty samples contained additional background DNA, while pure samples did not.

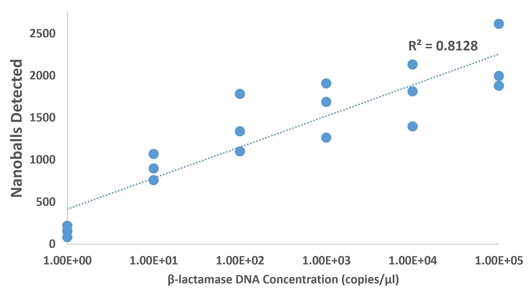

Figure 5. Number of nanoballs produced for increasing concentrations of target DNA gene block in samples amplified by the DNA nanoball kit.

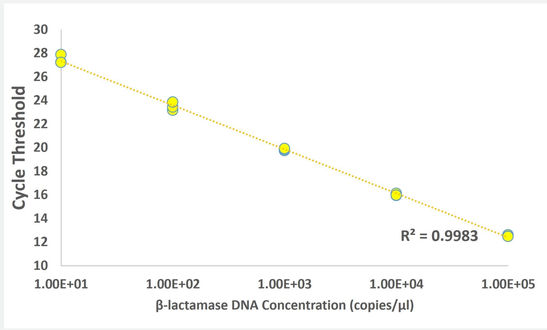

Figure 6. Cycle threshold numbers for increasing concentrations of target DNA gene block in dirty positive samples amplified through qPCR.

Discussion

- Positive samples transitioned from pink to yellow, negative samples did not for both dirty and pure conditions
- Methods were comparable in nanoball production for qualitative samples
- Both quantitative trials demonstrated a concentration curve

Future Directions

- This work was completed using a synthesized gene block for antibiotic resistance gene, next step is to develop targets directed for coral health
  - Areas of focus are for coral stress, disease presence, and reproduction cycle
- Goal is to test LAMP method with coral samples in the field

References

1. Rivera, H., Chan, A., & Luu, V. (2020). Coral reefs are critical for our food supply, tourism, and ocean health. We can protect them from climate change. MIT Science Policy Review. 1. 18-33. 10.38105/spr.7vn798jnsk.
2. Zhang, X., Zhao, Y., Zeng, Y., & Zhang, C. (2023). Evolution of the Probe-Based Loop-Mediated Isothermal Amplification (LAMP) Assays in Pathogen Detection. Diagnostics, 13(9), 1530. https://doi.org/10.3390/diagnostics13091530
3. Tayyab, M., Barrett, D., Van Riel, G., Liu, S., Reinius, B., Scharfe, C., Griffin, P., Steinmetz, L.M., Javanmard, M., Pelechano, V. (2023). Digital assay for rapid electronic quantification of clinical pathogens using DNA nanoballs. Sci. Adv., 9 (36), Article eadi4997, 10.1126/sciadv.adi4997