

Review of: "A Rapid and Robust DNA Extraction Method for PCR-Based Diagnosis of V. cholerae"

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Potential competing interests: No potential competing interests to declare.

Review report - Qeios- X0FQM7

Major Comments:

While you provide context about cholera and the limitations of conventional methods, some sections could benefit from a clearer structure. You could explicitly mention the knowledge gap that your study aims to fill (i.e., a rapid, broth-free DNA extraction for *V. cholerae* diagnostics).

Consider adding a comparison table in the introduction that highlights the time and cost differences between your method and conventional protocols like CTAB, phenol-chloroform, and broth culturing. This will better justify the novelty of your approach.

Minor Comments:

1. Materials and methods:

• Line: "Our method involves boiling of V. cholerae suspension in distilled water (takes 18-24 hours)."

Suggestion: "Our method involves boiling a V. cholerae suspension in distilled water, completing within 18-24 hours..."

• Line: "Rapid, simple, and less expensive molecular diagnosis has higher importance."

Suggestion: "Rapid, simple, and cost-effective molecular diagnostics are essential."

- 1. Make sure gene names (e.g., ctxA, tcpA) are italicized throughout the paper for consistency.
- 2. To improve readability and reproducibility, include a table summarizing PCR conditions (e.g., annealing temperatures, primer sequences, amplicon sizes). Clarify if **negative controls** were run with non-*V. cholerae* strains during the assays to demonstrate specificity.

3. Results:

• In the results section, you mention a figure showing PCR results ("Figure 1"), but this figure is missing from the text provided. Ensure figures are well-labeled and referenced properly. If gel electrophoresis images are provided, mention the molecular weight markers used.



4. References:

• Some references use inconsistent formats (e.g., [10], [11], [15]). Ensure all are correctly formatted according to the target journal's style.

5. Abstract Revision:

• The phrase "boiling of *V. cholerae* suspension in distilled water (takes 18-24 hours)..." could be misunderstood. Clarify that the 18-24 hour period includes the entire workflow, not just boiling.