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Peer Review

Review of: "Molecular Characterization of *Vibrio cholerae* Outbreak Isolates from Western India"

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This study provides a valuable molecular characterization of *Vibrio cholerae* isolates from a documented cholera outbreak in western India and contributes meaningful regional surveillance data on circulating atypical El Tor variants. The manuscript is methodologically structured, and the integration of PCR-based virulence gene profiling, sequencing confirmation, phylogenetic analysis, and protein-structure mutation mapping represents a notable strength. The identification of a Haitian-type ctxB7 genotype and its association with polymyxin B sensitivity adds epidemiological relevance and supports emerging reports describing evolutionary convergence between classical and El Tor phenotypes. Additionally, the detection of atypical enteropathogenic *Escherichia coli* suggests a potentially mixed microbial etiology, which enhances the public health significance of the investigation.

However, several limitations reduce the overall impact of the study. The very small sample size (n = 5 isolates) restricts epidemiological generalizability and limits the ability to assess intra-outbreak genetic diversity. The study would benefit from including environmental isolates or a larger patient cohort to strengthen outbreak source tracking and transmission analysis. The phylogenetic methodology relies solely on partial gene sequencing rather than whole-genome sequencing, which is now considered the gold standard for high-resolution cholera molecular epidemiology. Furthermore, antimicrobial susceptibility profiling is largely inferred from genetic similarity rather than experimentally validated phenotypic testing, which weakens conclusions regarding polymyxin sensitivity patterns. The functional implications of the TcpA N64S mutation remain speculative, and additional in vitro virulence or adhesion assays would significantly strengthen mechanistic interpretations. Finally, the discussion could better integrate global cholera genomic surveillance data to place the findings within a broader evolutionary context.

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Declarations

Potential competing interests: No potential competing interests to declare.