

Laboratory Detection of *Clostridium difficile* Infection

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Clostridium difficile infection (CDI) is caused by toxigenic strains of the organism that produces toxins A and B, with clinical disease requiring both the presence of significant diarrhea (≥ 3 stools per day) and laboratory detection of the toxin.¹ Rapid identification and treatment of infected patients is crucial, given the organism's highly transmissible nature through hardy spores and potential for causing pseudomembranous colitis.

Virulence of *C. difficile* is conferred by two exotoxins, toxin A and toxin B, encoded by their respective genes *tcdA* and *tcdB*. In addition, a hypervirulent strain of toxigenic *C. difficile* was identified in 2005.² This strain is characterized by the production of a binary toxin, encoded by the genes *ctdA* and *ctdB*.² While the exact role of the binary toxin in the disease caused by *C. difficile* remains unclear, epidemic strains (BI/NAP1/027) carrying this toxin show hypervirulence and behave synergistically with *tcdA* and *tcdB*, resulting in severe colitis.²

The traditional "gold standard" for the detection of CDI is the toxigenic culture, necessary for epidemiological studies but rarely used clinically due to its lack of standardization and four to seven-day turnaround-time (TAT). In contrast, enzyme immunoassays (EIAs), which detect toxin A and/or B are still widely used, despite their low sensitivities (30%–70%). Because of EIAs' known high false-positive rates, clinicians often order them on patients repeatedly, with sequentially lower positive predictive values.^{3,4} The Society for Healthcare Epidemiology of America and the Infectious Diseases Society of America (SHEA-IDSA) discourages the use of EIAs.⁵ An alternative method is a two-step algorithm: first detect GDH, a common antigen; then detect toxin with either EIA or cytotoxin neutralization assay.⁴

Since 2009, the US Food and Drug Administration (FDA) has approved several molecular-based tests with fast TAT and high sensitivities and specificities. These nucleic acid amplification assays include the PCR-based BD GeneOhm Cdiff assay (Becton Dickinson), proGastro Cd (Gen-Probe/Prodesse), and Xpert *C. difficile* (Cepheid). The Meridian illumigene *C. difficile* assay, which uses loop-mediated isothermal amplification to detect *tcdA*, is also FDA-approved. Currently, tests for the binary toxin are for research use only.

Laboratory considerations in assay selection include test accuracy, equipment and reagent costs, and technician time; but infection control and its impact on patient care are critical. Patients suspected of CDI must be placed on isolation precautions appropriate for those who are truly positive. Unfortunately, even false-positive test results will lead to patient isolation and treatment with oral vancomycin or metronidazole. Conversely, patients with false negative test results are not treated, potentially prolonging hospitalization with risk of transmission to hospital staff and other patients. Therefore, the indirect cost-savings of timely diagnoses and decreased transmission can offset assay costs.

A concerted effort by both pathologists and clinicians is needed to implement new tests and hospital policies successfully. Guidelines developed through collaboration can include the discouragement of "test for cure," which is not endorsed by SHEA-IDSA,⁵ as well as a mandatory waiting period. Though newer molecular tests might appear cost-prohibitive, tests that deliver results quickly and accurately may result in long-term cost savings and improved medical care.

References

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