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Microbial Stewardship (M Stevens, Section Editor)

Diagnostic Stewardship Approaches to *Clostridioides difficile* Infection in the Era of Two-Step Testing: a Shifting Landscape

Jennifer Emberger, MD, MPH[1,*]
Matthew M. Hitchcock, MD, MPH[1,2]
J. Daniel Markley, DO, MPH[1,2]

Address
[*,1]Department of Internal Medicine, Division of Infectious Diseases, Virginia Commonwealth University Medical Center, VMI Building, 2nd Floor, Room 205, Richmond, VA, 23298, USA
Email: jennifer.emberger@vcuhealth.org
[2]Department of Medicine, Division of Infectious Diseases, Hunter Holmes McGuire Veterans Affairs Medical Center, Richmond, VA, USA

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Abstract

Purpose of review To discuss the current strategies and impact of diagnostic stewardship for *Clostridioides difficile* infection.

Recent findings The diagnosis of *C. difficile* infection is challenging due to complex epidemiology and the limitations of a single assay that is adequate for diagnosis. Over-diagnosis with sensitive molecular assays is common due to the prevalence of colonization with *C. difficile*. To overcome these challenges, multiple diagnostic stewardship strategies have been successfully deployed to optimize *C. difficile* testing.

Summary Diagnostic stewardship strategies should be implemented at every stage of *C. difficile* testing in order to limit testing to patients with a high pre-test probability, minimize the limitations of stand-alone assays, and guide clinicians to appropriate management through clear result reporting and interpretation.

Introduction

While pseudomembranous colitis (PMC) was first identified histopathologically in the 1890s and *Clostridioides* (previously *Clostridium*) *difficile* was isolated as a colonizing organism in infants in 1935, the two were not linked until the late 1970s when PMC was revealed to be a toxin-mediated *Clostridial* process [1–3]. Today, *C. difficile* infection (CDI) is considered one of the five urgent public health threats included in the Centers for Disease Control and Prevention's (CDC) *Antibiotic Resistance Threats in the United States 2019* report [4]. In 2017, there were approximately 224,000 cases of CDI in hospitalized patients and 12,800 deaths [4]. In 2020, the diagnosis of CDI can be made using a variety of tests, but due to limitations in diagnostic accuracy and an evolving understanding of the degree of *C. difficile* carriage within patient populations, the optimal diagnostic strategy remains undefined [5–7]. Unfortunately, no validated diagnostic criteria for CDI exist at this time. As our understanding of CDI epidemiology and diagnostic limitations expands, the role of diagnostic stewardship has become increasingly relevant. Diagnostic stewardship is an emerging term for the careful use of diagnostics in order to focus testing toward higher-probability settings, especially when diagnostics are highly sensitive but lack specificity, thus increasing the likelihood that results are clinically meaningful [8–10]. The most recent Infectious Diseases Society of America (IDSA)/Society for Healthcare Epidemiology of America (SHEA) and European Society of Clinical Microbiology and Infectious Diseases (ESCMID) guidelines for the management of CDI advise a blend of algorithmic diagnostic strategies and diagnostic stewardship [5, 11–13].

Herein, we describe the current state of diagnostic stewardship for CDI, including strategies to optimize the patient population tested (pre-analytic), maximize the sensitivity and specificity of the available diagnostic tests (analytic), and clarify result reporting (post-analytic) in order to guide clinicians to appropriate management.

Evolution of CDI diagnostic testing

Cell cytotoxicity neutralization assay (CCNA) and toxigenic culture

In order to understand the diagnostic challenges of testing for CDI, it is critical to recognize the historical evolution of testing strategies and their diagnostic characteristics (Table 1). The first diagnostic test developed was CCNA, which evaluated for the presence of free *Clostridial* toxin by applying filtered stool to a fibroblast cell line in both the presence and absence of antitoxin in order to observe a cytopathic effect that is neutralized by antitoxin over a 24–48 h period [14]. While the CCNA has high specificity and remains the analytical gold standard for toxin-mediated CDI [15], the process is time-, labor-, and resource-intensive, and difficult to standardize. Since the 1980s, it has been well recognized that the poor sensitivity of CCNA runs the risk of missing cases of CDI [16]. Toxigenic culture, which is the most sensitive test for isolation of toxin-producing *C. difficile*, is even more labor intensive than CCNA, but is known to lack specificity (17).

Enzyme immunoassays (EIA) and glutamate dehydrogenase (GDH)

The intense resource requirements and diagnostic limitations of CCNA and toxigenic culture led to the development of EIA using antibodies to detect free *Clostridial* toxins A and B, as well as GDH, an enzyme produced and secreted at high levels by all *C. difficile* strains (both toxigenic and non-toxigenic) [18]. Unfortunately, these toxin EIAs were found to lack sensitivity, which led to the institutional practice of performing serial toxin EIAs on patients in order to increase the sensitivity [19]. While direct toxin assays have high sensitivity compared to CCNA [20], the sensitivity is much lower compared to toxigenic

Table 1. Comparison of available tests for the diagnosis of *C. difficile* infection

Diagnostic Test	Strength	Weakness
Cell cytotoxicity neutralization assay (CCNA)	• High specificity • Analytical gold standard for toxin-mediated CDI	• Resource-intensive • Technically complex • Difficult to standardize
Toxigenic culture	• Highest sensitivity for presence of toxigenic *C. difficile*	• Resource-intensive • Technically complex • Low specificity (cannot distinguish between colonization and toxin-mediated disease)
Enzyme immunoassays (EIA) for free toxin	• High specificity • Rapid turnaround time • Simple, low-cost test	• Low sensitivity
Glutamate dehydrogenase (GDH)	• High sensitivity • Rapid turnaround time • Simple, low-cost test	• Low specificity (cannot distinguish between toxigenic and non-toxigenic strains of *C. difficile*)
Nucleic acid amplification t(NAAT)	• High sensitivity for presence of toxigenic *C. difficile* • Rapid turnaround time	• Low specificity (cannot distinguish between colonization and toxin-mediated disease)

culture, and they were largely abandoned as stand-alone tests due to concerns about missed cases [21–24]. GDH assays are highly sensitive for detection of *C. difficile*; therefore, they have a high negative predictive value and are helpful as an initial screen for ruling out infection [7]. Conversely, GDH has a lower positive predictive value due to its inability to discriminate between toxigenic and non-toxigenic *C. difficile* strains and must be paired with a test that detects toxin or toxigenic potential [7, 25, 26].

Nucleic acid amplification tests

The next major development in CDI diagnostic testing was catalyzed by the emergence of the hypervirulent NAP-1/BI/027 strain of *C. difficile*, which conferred higher morbidity and mortality [27]. In this context, concerns over false negative toxin EIAs renewed interest in high-sensitivity testing. Nucleic acid amplification tests (NAATs), primarily using polymerase chain reaction (PCR), were developed, and these new tests offered sensitivity comparable to toxigenic culture with results available in as little as an hour [22, 28, 29]. However, because NAATs detect genes encoding for toxin and not toxin itself, they cannot distinguish between colonization and toxin-mediated disease, especially in the absence of clinically significant diarrhea [30]. The switch to these highly sensitive molecular tests resulted in a large increase in case number (40–100% in some laboratories) without clear evidence of heightened transmission or increased antibiotic use, suggesting that NAATs were detecting not only cases missed by toxin testing, but also patients with subclinical disease or colonization [31–36].

Evolving understanding of CDI transmission

Detection of colonized patients has facilitated a paradigm shift in our understanding of *C. difficile* transmission. While CDI has historically been considered primarily a nosocomial infection, more recent studies have shown that nosocomial acquisition may account for less than a third of CDI cases [37–39]. Additionally, it seems that most cases of CDI are acquired from sources other than symptomatic cases [37, 40]. Studies have shown that up to 15% of healthy individuals in the community and up to 20% of hospitalized patients are colonized with *C. difficile* [6]. Although the role of asymptomatic carriage in the transmission of CDI is not entirely known, there is evidence to support an increased risk of CDI with exposure to asymptomatic carriers [41, 42].

The role of diagnostic stewardship

Amidst this potpourri of diagnostic testing with variable test characteristics, several key studies highlighted the importance of incorporating clinical symptoms into assay interpretation and the critical role of diagnostic stewardship. A study by Dubberke et al. evaluating nine *C. difficile* diagnostic assays observed that NAATs had the lowest specificity and significant overutilization was occurring, as 36% of patients did not have clinically significant diarrhea at the time of testing and 20% were receiving laxatives [30]. Planche et al. sought to validate CDI tests based on clinical outcomes and compared four commercial assays including two toxin EIAs, GDH, and NAAT to the reference standards (CCNA and toxigenic culture), and observed that CCNA-positive patients had more severe disease and higher mortality compared to PCR-positive/toxin-negative patients or CCNA-negative patients [15]. While CCNA had the best diagnostic accuracy for CDI, the turnaround time for the method was unacceptable, and multi-step diagnostic algorithms including GDH and sensitive toxin EIA tests were recommended [15]. In this context, researchers have sought to determine whether PCR-positive/toxin-negative patients actually require treatment. Polage et al. addressed this question by comparing patients managed by clinically reported toxin EIA results to those who were positive only by NAAT performed for research purposes and not reported to clinicians, and observed that PCR-positive/toxin-negative patients had shorter duration of symptoms and milder clinical disease with outcomes similar to patients negative by all modalities, and 95% of the CDI-related deaths and complications occurred exclusively in toxin-positive patients [43•]. These key studies raised significant concerns of overdiagnosis of CDI when using stand-alone NAAT testing.

Finding the "sweet spot" for CDI testing

Given the weaknesses of each diagnostic approach, finding the so-called "sweet spot" for *C. difficile* diagnostic testing that minimizes overdiagnosis without missing cases cannot be achieved with stand-alone testing. Diagnostic stewardship strategies are critical in order to minimize the effects of assay limitations and optimize appropriate testing. The 2017 IDSA/SHEA Clinical Practice Guidelines for *C. difficile* Infection in Adults and Children recognize the importance of diagnostic stewardship by recommending that institutions establish pre-agreed upon criteria for appropriate sample submission for *C. difficile*

testing in order to minimize stand-alone diagnostic limitations [11••]. The guidelines suggest that the most appropriate patient population for testing are those with unexplained, new-onset diarrhea with ≥3 unformed stools within 24 h. If institutional criteria are not in place, it is recommended that multi-step testing be performed [11••]. This guideline underscores the dynamic interplay between diagnostic stewardship and testing methodology.

Diagnostic stewardship opportunities with *C. difficile*

The overarching goal of antimicrobial stewardship is to optimize the appropriate use of antimicrobials and reduce overuse when antibiotics are not warranted. Similarly, the goal of diagnostic stewardship is to optimize the appropriate use of diagnostic testing and reduce overuse when testing is not warranted. There are three moments of diagnostic stewardship that correspond to the stages of diagnostic testing, namely pre-analytic, analytic, and post-analytic [13] (see Table 1). The pre-analytic stage refers to the decision to test and the choice of appropriate specimens for testing, the analytic stage refers to how the test is performed, and the post-analytic stage refers to how the results are represented and interpreted [13]. Within each of these stages, various strategies have been utilized including education, pre-authorization of testing, prospective audit and feedback, computerized clinical decision support (CCDS), and other approaches (Table 2).

Pre-analytical diagnostic stewardship strategies

Stool rejection criteria

The pre-analytical phase is a critical time to optimize *C. difficile* testing by refining the population tested in order to maximize the likelihood that a positive test corresponds to a true clinical diagnosis of CDI [8, 44]. The most common and simple example of this strategy is laboratory-based stool rejection criteria. Here, the laboratory denies testing formed stools for *C. difficile*, as patients producing formed stools are highly unlikely to have a clinical diagnosis of CDI [45]. This is the simplest pre-analytic strategy to implement as it only involves assessment of the submitted stool sample by laboratory personnel prior to testing and does not require alteration of physician ordering behavior. This approach is recommended in the most recent IDSA/SHEA and ESCMID guidelines [5, 11]. More complex rejection criteria have also been described which deny testing in various scenarios including: negative *C. difficile* NAAT within 7 days (as repeat testing in this interval is highly unlikely to change the result) [45, 46], recent laxative administration (often within 24–48 h), and/or nursing documentation of <3 diarrheal stools within a 24 h period [46, 47]. Truong et al. found that applying strict pre-analytic criteria reduced *C. difficile* testing and hospital-onset *C. difficile* infection [HO-CDI] rates by 30% with no association with increased complications in patients with canceled orders [47•]. It should be noted that criteria of higher complexity require an electronic medical record (EMR) that can provide the necessary data, accurate documentation of stool output by nursing, and significant information technology support. While rejection criteria based on recent laxative administration is beneficial, this

Table 2. Diagnostic stewardship strategies stratified by stage of testing

Level and Type of Intervention	References
Pre-Analytical	
Laboratory Rejection Criteria - Formed Stools - Recent Laxative Administration - Recent *C. difficile* Test	Kraft et al. [45] Luo et al. [46] Truong et al. [47•]
Provider Education - Institutional Testing Criteria - Educational Presentations to Front-line Clinicians	Yen et al. [52] Kociolek et al. [53] Christensen et al. [54•] Turner et al. [100] Thompson et al. [101]
Computerized Clinical Decision Support Tools at Order Entry - Recent *C. difficile* Test Alert - Recent Laxative Administration Alert - Attestation of ≥3 Diarrheal Stools in 24 hours - Institutional Testing Criteria Displayed	Kociolek et al. [53] Christensen et al. [54•] Luo et al. [55] Bilinskaya et al. [56] White et al. [57] Kwon et al. [58] Howard-Anderson et al. [59] Quan et al. [60] Fleming et al. [62] Madden et al. [636] Friedland et al. [64] Mizusawa et al. [65] Turner et al. [100] Sperling et al. [61] Cook et al. [102]
Test Restriction with Pre-Approval - Sub-specialty physician approval - Laboratory staff approval	Kwon et al. [58] Quan et al. [60] Mizusawa et al. [65] Lin et al. [67] Drees et al. [103]
Post-Order Prospective Audit and Feedback - Review of indications for testing	Christensen et al. [54•] Jakharia et al. [68]
Analytical Interventions	
Multi-Step Algorithms PCR Cycle Threshold to Predict Toxin Positivity	Crobach et al. [5••] McDonald et al. [11••] Gateau et al. [70] Hitchcock et al. [82] Orendi et al. [104]
Post-Analytical Interventions	
Toxin-Dominant Reporting Interpretive Comments in Result Report	Planche et al. [15] Polage et al. [43•] Hitchcock et al. [82] Schwenk et al. [85] Polage et al. [105]

approach has the potential to delay diagnosis of CDI as laxative use does not preclude the possibility of CDI. Laxatives are commonly given to inpatients and have a wide spectrum of potency, and often, patients have been receiving laxatives long before developing acute diarrhea [48]. A recent study found no difference in the rate of severe CDI by objective scoring methods between patients with a positive *C. difficile* NAAT who had received laxatives and those who did not [49]. This finding underscores the importance of acknowledging that patients on laxatives can still develop CDI.

Education

Provider education is a common pre-analytic strategy to improve ordering behavior, though this is generally considered a less effective and sustainable intervention unless combined with other methods [50, 51]. Two recent studies that successfully used didactic education as a core strategy paired this approach with introduction or reinforcement of laboratory rejection criteria and offered ongoing provider feedback on appropriate testing using a hospital-wide educational screensaver or an alert within the EMR at the point of testing [52, 53]. Another study paired education with decision support at order entry and post-order prospective audit and feedback (PAF) in certain contexts with varying success [54•].

Computerized clinical decision support (CCDS)

Implementing computerized clinical decision support (CCDS) at the time of order entry for *C. difficile* testing is a common strategy to increase the appropriateness of testing. This pre-analytic intervention runs continuously with limited maintenance following initial development. Decision support tools of varying complexity have been described, and examples range from simple alerts of a recent negative *C. difficile* test result or receipt of a laxative [55–59] to more complex ones with multiple steps that may require ordering clinicians to affirm that patients have indeed had ≥3 liquid stools within 24 h and have not recently received laxatives and/or that specific signs or symptoms of CDI are present without an alternative cause of diarrhea [54, 60–65]. While CCDS tools are common, high rates of alert override have been noted and concerns about alert fatigue and loss of clinical autonomy have been described in qualitative research [56, 65, 66]. Other approaches, generally used in tandem with CCDS and laboratory rejection criteria, are more resource-intensive and include order restriction requiring infectious diseases or laboratory approval [58, 65, 67] and post-order PAF [54, 68].

Analytical diagnostic stewardship strategies

Analytical diagnostic stewardship strategies address optimization of the testing approach for the diagnosis of CDI. The 2017 IDSA/SHEA guidelines define testing approaches into two main categories: NAAT alone with pre-agreed upon institutional guidelines for appropriate specimen submission or a multi-step algorithm with or without pre-agreed upon institutional guidelines [11••]. There are strengths and weaknesses to each approach. Pre-and post-analytic strategies can be utilized to minimize the weaknesses of each analytic approach

and optimize the clinical relevance of the results. Pre-analytic diagnostic testing strategies can be applied to any diagnostic testing approach and are not affected significantly by choice of testing modality. However, post-analytic strategies will differ by testing modality as the way a test is reported and interpreted is dependent on which test(s) are utilized.

NAAT alone with pre-agreed upon institutional guidelines

The key strengths of NAATs are the rapid availability of results coupled with a high sensitivity and negative predictive value for the presence of toxigenic *C. difficile* [25, 26, 69]. However, NAATs cannot distinguish between patients who have CDI and patients who are merely colonized with toxigenic *C. difficile*, leading to overdiagnosis and unnecessary treatment [43, 69, 70]. Unnecessary treatment with oral vancomycin is not without risk and has been associated with significant shifts in the gut microbiome, variable recovery time after cessation of therapy, and predisposition to colonization with resistant organisms [71, 72]. Overdiagnosis is also associated with increased laboratory-identified (LabID) HO-CDI events that are reported to the National Healthcare Safety Network (NHSN) which can have punitive financial implications for hospitals [73, 74].

Pre-analytic diagnostic stewardship strategies can help optimize stand-alone NAAT testing. Pre-agreed upon criteria for appropriate specimen submission helps assure that the right patient population is being tested [11••]. Most importantly, testing should only be performed in patients with new-onset diarrhea, defined as ≥3 unformed stools in a 24-h period without a clear alternative etiology [11••]. A variety of pre-analytic diagnostic stewardship strategies have been studied to reduce potentially inappropriate *C. difficile* testing when PCR assays are used as stand-alone tests (Table 2). A small network of hospitals introduced an alert embedded in the EMR that fired when *C. difficile* testing was ordered on a patient who had received laxatives in the past 24 h and recommended canceling the test, which was implemented alongside pre-existing laboratory rejection of formed stools submitted for testing [56]. The alerts were only modestly successful as 75% were immediately overridden, and testing was re-ordered within 24 h on 13% of initially accepted alerts [56]. Christensen et al. reported the use of a multi-tiered approach that included laboratory criteria requiring liquid stool for testing, provider education on appropriate testing context, a CCDS within the EMR, and Antibiotic Stewardship Program (ASP) pre-authorization of testing on or after the fourth day of a patient's hospitalization, with providers allowed to accept or reject the recommendation [54•]. This intervention was associated with a significant reduction in positive NAATs per month, HO-CDI rate, and oral vancomycin consumption [54•]. Sperling et al. also utilized a multi-tiered approach with standardization of laboratory criteria for testing, implementation of CCDS at order entry, and PAF by infection preventionists regarding appropriateness of testing, and this bundle was associated with a 40% reduction in testing, 59% reduction in HO-CDI rates, and a 27% reduction in oral vancomycin usage [61]. Lin et al. found that their CCDS intervention was often disregarded and implemented mandatory ID physician approval of *C. difficile* testing performed on or after hospital day 4, which reduced ordering volume by >50% and HO-CDI rates by 40% [67].

Post-analytic interventions with a stand-alone NAAT testing should provide educational support for interpretation of positive PCR results that reinforce that colonization is common and clinical context is key to determining whether a patient truly has CDI [53].

PCR cycle threshold reporting

Typically, stand-alone PCR testing is reported as binary, qualitative result (positive or negative). The number of PCR cycles required to turn an assay "positive" is known as the PCR cycle threshold (CT). Studies have observed that the PCR CT value is inversely correlated with organismal load [75, 76], and patients with lower CT values tend to have more severe disease and a higher risk of recurrence [77–82]. PCR CT has been used to predict toxin positivity by EIA [6, 83, 84], and this methodology maintains the sensitivity of NAAT while producing additional information about the likelihood of toxin-mediated disease without a second test. Studies have revealed that PCR CT may help discriminate between patients more likely to have CDI and those who are colonized and do not require therapy [82, 85]. However, there is significant overlap in CT values between toxin EIA-positive and toxin EIA-negative patients, which lowers the specificity [79, 86]. This methodology is only in limited use and requires local validation of a cut-point and ongoing quality control evaluation.

Algorithmized combination testing with/without pre-agreed upon institutional guidelines

Both the IDSA/SHEA and ESCMID guidelines promote multi-step algorithmized testing for CDI [5, 11]. There are essentially three main options for multi-step algorithmized testing: (1) GDH followed by toxin EIA, (2) GDH followed by toxin EIA, with NAAT testing performed when GDH and toxin EIA are discordant (primarily GDH+/Toxin−), and (3) NAAT followed by toxin EIA.

GDH followed by toxin EIA

Because of the limitations of toxin EIAs and GDH assays alone, these assays are paired together to optimize their complementary strengths; the high sensitivity of GDH assays reduces the risk of false negatives and the toxin EIAs identify patients more likely to have true, toxin-mediated CDI that requires therapy [7, 26]. This combination algorithm tends to be cheaper than stand-alone NAATs, and rapid EIAs for both toxin and GDH in a single test are available [25, 87]. The major weakness of this algorithm is the commonly encountered discordant result (GDH+/Toxin−), which may be due to presence of a non-toxigenic strain, presence of toxigenic organisms in a colonizing state, or a false negative toxin assay (given the lower sensitivity of these tests) [15, 25, 26].

GDH followed by toxin EIA, arbitrated by NAAT for discordant results

When results of the GDH followed by toxin EIA algorithm are discordant (GDH+/Toxin−), a third step, generally a NAAT, can be added to help differentiate between a non-toxigenic and a toxigenic strain and increases the overall sensitivity of the algorithm for presence of toxigenic *C. difficile* [7, 25, 26]. However, the inherent limitation of NAATs being unable to differentiate between colonization and toxin-mediated disease remains, so this multi-step algorithm must still be interpreted with caution as a positive NAAT following discordant GDH and toxin results is not diagnostic of CDI.

NAAT followed by toxin EIA

An alternative two-step algorithm leads with a NAAT and reflexes to toxin EIA if the NAAT is positive. This takes advantage of the high sensitivity and negative predictive value of NAATs, allowing CDI to effectively be ruled out if negative. The reflex to a toxin EIA uses the high specificity of toxin tests, meaning that NAAT+/Toxin+ patients are highly likely to have true CDI. As with the prior multi-step algorithm, interpretation of NAAT+/Toxin– patients remains difficult, as this is a heterogeneous population, with some of these patients having CDI with a false negative toxin result that requires treatment, but many are likely to have colonization [88, 89].

Post-analytical diagnostic stewardship strategies

Post-analytical diagnostic stewardship strategies refer to how results are reported and what interpretive support is offered. Multi-step algorithms introduce myriad options for reporting, and there is a paucity of data to guide optimal strategies for reporting algorithmized combination results. Each step can be reported out individually, which leaves the overall interpretation of the combination of results up to the ordering provider. This approach would require a sophisticated understanding of *C. difficile* epidemiology and the meaning of individual tests by frontline clinicians. Given the complexities with individual component result reporting, the ASP, microbiology laboratory, and frontline clinicians should work together to develop a reporting approach that minimizes confusion and accounts for the various permutations of results. Interpretive comments should always include a recommendation prompting providers to take clinical context into account. As Dubberke and Burnham aptly state, "it is best to remember to treat the patient, not the test." [90]

Alternatively, the results of multi-step algorithms can be reported in a binary fashion (positive or negative) based on the last test performed in the algorithm. While this approach does decrease the likelihood of inter-user variability in result interpretation, it does not account for the complexities of *C. difficile* diagnostics. Evidence from the use of stand-alone PCR suggests that there is a bias toward treatment when a positive result is reported, regardless of clinical context [13, 51, 82]. Moreover, binary reporting deprives the frontline provider of knowing about the possibility of colonization (NAAT+/Toxin-), which could have implications for treatment and transmission-based precautions. From an epidemiologic perspective, there is evidence that asymptomatic carriers often have skin and environmental contamination with *C. difficile* [91, 92], as well as an increased risk of CDI with exposure to asymptomatic carriers [41, 42]. However, contact precautions can have negative impacts on patient care and patient experience and require careful consideration, especially with regard to placement of asymptomatic carriers on isolation [93, 94]. This conundrum highlights the dynamic interplay between infection control and diagnostic stewardship.

Toxin-dominant reporting

With the growing body of literature supporting the association between toxin positivity and risk of severe CDI and adverse outcomes [15, 43, 95], there seems to be a general trend toward toxin-dominant reporting. Patients who are toxin-

positive are thought to have a "positive" result for CDI and those who are toxin-negative either are "colonized" or have mild CDI that may not require therapy. Toxin-dominant reporting has advantages over stand-alone NAAT testing even with pre-agreed institutional guidelines for stool submission, as the latter is fraught with issues of overdiagnosis and subsequent overtreatment. Toxin-dominant reporting may become standardized over time as CDI toxin testing continues to evolve and improve. The utilization of PCR cycle threshold (CT) to predict toxin assay positivity and the development of ultrasensitive toxin assays could also provide significant opportunities for diagnostic stewardship in the future [6, 82, 83, 96, 97].

Pitfalls of diagnostic stewardship strategies

It is important to consider that even small diagnostic stewardship interventions may cause a cascade of downstream effects within a health system. Draconian pre-analytic strategies may lead to unintended consequences. Caution is advised when applying so-called "hard stops" in CCDS pathways, as patients on laxatives can still develop CDI and CCDS should not override appropriate clinical judgment. Over-restriction of testing may lead to missed diagnoses, so it is prudent to ensure providers have recourse to contact the laboratory or ASP if needed to justify testing when indicated. Pre-authorization of *C. difficile* testing or CCDS interventions that only activate after day 3 of hospitalization can cause credibility issues for an ASP, as this approach gives the impression that it is only important to optimize testing when the hospital is at risk for an NHSN LabID event and financial penalties. Optimization of testing should be emphasized across the continuum of care, regardless of when or where the test is ordered. Algorithmic approaches to testing may be associated with increased costs, labor, and prolonged turnaround times. As noted above, algorithms also introduce the possibility of confusion when reporting results, as discordant results (NAAT+/Toxin−) require careful interpretation. Frontline providers should be assisted with interpretation of these results and the decision to treat versus observe a patient. Infection control surveillance is also impacted, as best practices for transmission-based precautions for NAAT+/Toxin− patients remain unclear at this time. Longitudinal feedback and tracking of performance metrics by the ASP are critical whenever a new diagnostic stewardship strategy is implemented. It should be noted that multiple diagnostic stewardship strategies are often employed simultaneously, making it difficult to truly determine the most effective approach.

Conclusions

Given the complexities of *C. difficile* epidemiology and the lack of a definitive diagnostic standard with the current generation of assays, diagnostic stewardship is critical to ensure that the right patients are tested and correct CDI diagnoses are made so that treatment can be administered appropriately and unnecessary antibiotics avoided [10, 98, 99]. In many ways, diagnostic stewardship is an upstream form of antimicrobial stewardship, as a refinement in diagnosis leads to better usage of antibiotics [10]. While strategies can target any stage of testing, interventions would ideally include the pre-analytical phase, as

choice of specific algorithms or assays cannot take the place of properly selecting the patient population being tested. Numerous pre-analytical interventions have been successfully implemented; nonetheless, this remains an area of intense research. Analytical phase interventions are also critical, as multi-step algorithms can help differentiate between toxin-mediated CDI and colonization better than stand-alone NAAT [15], and lead to more appropriate therapy decisions. However, there is still much to understand about patients with discordant results. Though these patient are likely to be colonized, a proportion certainly have true CDI, and while multiple recent studies have evaluated this population in a retrospective fashion [82, 88, 89], more studies, especially prospective, are needed to better determine who in this population benefits from treatment. Patients with discordant results also offer an opportunity for post-analytical antimicrobial stewardship interventions to help frontline clinicians interpret multi-step algorithm results, determine whether therapy is indicated, and choose guideline concordant treatment. Until the promise of a single test that combines both high sensitivity and specificity is fully realized, diagnostic stewardship will remain an indispensable strategy in the diagnosis of CDI.

Compliance with Ethical Standards

Conflict of Interest

Jennifer Emberger declares that she has no conflict of interest.
Matthew Hitchcock declares that he has no conflict of interest.
J. Daniel Markley declares that has no conflict of interest.

Human and Animal Rights and Informed Consent

This article does not contain any studies with human or animal subjects performed by any of the authors.

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- • Of importance
- •• Of major importance

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