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Early Diagnosis, Prevention, and Treatment of C. difficile: Update

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Preface

The Agency for Healthcare Research and Quality (AHRQ), through its Evidence-based Practice Centers (EPCs), sponsors the development of systematic reviews to assist public- and private-sector organizations in their efforts to improve the quality of health care in the United States. These reviews provide comprehensive, science-based information on common, costly medical conditions, and new health care technologies and strategies.

Systematic reviews are the building blocks underlying evidence-based practice; they focus attention on the strength and limits of evidence from research studies about the effectiveness and safety of a clinical intervention. In the context of developing recommendations for practice, systematic reviews can help clarify whether assertions about the value of the intervention are based on strong evidence from clinical studies. For more information about AHRQ EPC systematic reviews, see www/effectivehealthcare.ahrq.gov/reference/purpose.cfm.

AHRQ expects that these systematic reviews will be helpful to health plans, providers, purchasers, government programs, and the health care system as a whole. Transparency and stakeholder input are essential to the Effective Health Care Program. Please visit the Web site (www.effectivehealthcare.ahrq.gov) to see draft research questions and reports or to join an e-mail list to learn about new program products and opportunities for input.

We welcome comments on this systematic review. They may be sent by mail to the Task Order Officer named below at: Agency for Healthcare Research and Quality, 540 Gaither Road, Rockville, MD 20850, or by email to epc@ahrq.hhs.gov.

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Early Diagnosis, Prevention, and Treatment of C. difficile: Update

Structured Abstract

Objective. To update a 2011 review of differences in accuracy of diagnostic tests and the effects of interventions to prevent and treat C. difficile infection (CDI) in adults.

Data sources. Medline, the Cochrane Clinical Trials Registry, EMBASE, from 2010 through July 2014 plus reference lists of included studies and recent systematic reviews.

Methods. Two investigators screened abstracts and full texts of identified references for eligibility. Eligible studies included studies of sensitivity and specificity for diagnostic tests in patients at risk for CDI. We included randomized controlled trials or high quality cohort studies enrolling adult patients with CDI or suspected CDI for treatment interventions. Prevention studies also included adult patients at risk for CDI and observational study designs. Two investigators extracted data, assessed risk of bias on individual studies, and evaluated strength of the body of evidence for each comparison and outcome. Pooled estimates were analyzed to assess the efficacy and comparative effectiveness of a variety of treatments.

Results. We identified 31 diagnostic studies and 48 studies evaluating prevention or treatment interventions to update the review. High-strength evidence showed nucleic amplification tests were sensitive and specific for CDI. Low-strength evidence found some institutional prevention interventions, such as antibiotic prescribing practices and transmission interruption (terminal room cleaning with hydrogen peroxide vapor, bathing patients with chlorhexidine gluconate, handwashing campaigns) reduces CDI incidence. Low-strength evidence also suggests prevention interventions can be sustained over several years. For treatment of CDI, vancomycin is more effective than metronidazole (high-strength evidence), and the effect does not vary by severity (moderate-strength evidence). Fidaxomicin remains noninferior to vancomycin for the initial cure of CDI (moderate-strength evidence), but is superior to vancomycin for prevention of recurrent CDI (now high-strength evidence). Despite identifying 29 new studies for fecal microbiota treatment (FMT) or probiotics, evidence remains low-strength for FMT and lactobacillus strains being more effective than placebo for reducing recurrent CDI; probiotics using multiple organisms or Saccharomyces boulardii were not more effective. Evidence for FMT for refractory CDI was insufficient. Few studies reported adverse events; when reported, few events were noted.

Conclusions. Research on diagnostic testing for and interventions to treat CDI expanded considerably in 4 years. Nucleic acid amplification tests have high sensitivity and specificity for CDI. Vancomycin is more effective than metronidazole for initial CDI, while fidaxomicin is more effective than vancomycin for the prevention of recurrent CDI. FMT and lactobacillus probiotics to restore colonic biodiversity and improve patient resistance to CDI or recurrence have low strength but relatively consistent positive evidence for efficacy.

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Introduction

Condition

Clostridium difficile infection (CDI) rates in the United States and the world have increased in the last decade, along with associated morbidity and mortality. *Clostridium difficile* is a gram-positive, anaerobic bacterium generally acquired through ingestion. Various strains of the bacteria may produce disease generating enterotoxin A and cytotoxin B, as well as the lesser understood binary toxin. Our use of the term CDI indicates this review's focus is the presence of clinical disease rather than asymptomatic carriage of *C. difficile*. CDI symptoms can range from mild diarrhea to severe cases including pseudomembranous colitis and toxic megacolon and death. The estimated CDI mortality rate was 2.4 deaths per 100,000 population in 2011.[1,2] Beginning in 1999, the CDI mortality rate rose each year to a peak of 2.4 deaths per 100,000 in 2008, then decreased to 2.2 in 2009 and 2010 before rising again to 2.4 in 2011.

Distribution of CDI in the population is bimodal, with the largest incidence in older adults, who also experience the vast majority of severe morbidity and mortality. A much smaller peak occurs in children under age 10.[3,4] In 2011, 93 percent of CDI deaths occurred in individuals ≥65 years of age; CDI was the 17th leading cause of death in this age group.[2] Up to half of health-care associated CDI cases begin in long-term care, thus residents in these facilities are at high risk.[5] Incidence rates may increase by four- or five-fold during outbreaks.[6] Community-acquired and community-onset CDI, where CDI occurs outside the institutional setting, is also on the rise, though still generally lower than institution-acquired rates, accounting for 27 percent of cases in a recent prevalence study,[7] Community-associated CDI complicates measuring the effectiveness of prevention within an institutional setting.[6] Additionally, the pathogenesis of CDI is complex and incompletely understood, and onset may occur as late as several months after hospitalization or antibiotic use.

New, hypervirulent *C. difficile* strains have emerged since 2000. These affect a wider population that includes children, pregnant women, and other healthy adults, many of whom lack standard risk profiles such as previous hospitalization or antibiotic use.[8] The hypervirulent strains account for 51 percent of CDI, compared to only 17 percent of historical isolates.[9,10] Time from symptom development to septic shock may be reduced in the hypervirulent strains, making quick diagnosis and proactive treatment regimens critical for positive outcomes.

Diagnosis

Effective containment and treatment of CDI depends on accurate and swift diagnosis. An increasing number of diagnostic tests are designed to detect either the presence of the organism or toxins A and/or B with a variety of sensitivities, specificities, predictive values, biotechnologies used, training required, costs, and time-to-results. The testing strategies used in health systems are rapidly evolving. A study from 2008 showed that more than 90 percent of labs in the United States use enzyme immunoassay because it is fast, inexpensive, and easy to perform.[11] Just 3 years later, however, data showed that 43 percent of laboratories in the United States used nucleic acid amplification tests (NAAT) (e.g., polymerase chain reaction [PCR]).[12]

Clinically, CDI is diagnosed using tests such as: 1) immunoassays (including enzyme immunoassays, enzyme-linked immunosorbent assays, and immunochromatography assay), 2) tests for *C. difficile* toxins, and 3) amplification of *C. difficile* DNA, through means such as PCR and loop mediated isothermal amplification (LAMP). Some diagnostic testing strategies rely on

two-step procedures, the first being a sensitive, inexpensive, fast screen for the presence of the organism and, if that is positive, a second test for toxins. Toxigenic culture and cell cytotoxicity neutralization assay are no longer standard practice and are not universally available. However, given the rapid evolution of testing strategies, studies of diagnostic test performance often use toxigenic culture or cell cytotoxicity neutralization assay as the reference standard. Clinicians are not always well informed on the best diagnostic test to use, the operating characteristics of the tests used in their practice setting, or the relatively low likelihood of a false negative result (e.g., evidence suggests retesting with the same test is common practice, yet not recommended).

Treatment Strategies

Treatment for mild to moderate CDI is generally metronidazole, in part because of concerns that overuse of vancomycin may contribute to increasing pathogen resistance. Vancomycin is recommended for severe initial incident CDI.[13] However, both vancomycin and metronidazole have been implicated in leading to increased frequency of vancomycin-resistant enterococci.[14] In 2011, the FDA approved a new agent, fidaxomicin, for the treatment of CDI. A previous review found that while fidaxomicin was not superior for the initial cure of CDI, recurrence was less frequent with fidaxomicin than with vancomycin.[15] Measuring cure, however, can be challenging; no specific consensus exists regarding symptom resolution, clearance of the organism, or recurrence of CDI.

Treatment for relapsed or recurrent CDI is even more problematic. CDI recurs in 15 – 35 percent of patients with one previous episode and 33 – 65 percent of patients with more than two episodes.[16] Currently, clinicians choose from a number of antibiotics, dosing protocols, and adjunctive treatments (such as the use of antimicrobials, probiotics, toxin-binding agents, and immune-system enhancing agents).[17-19] The goal of most adjunctive treatments is to reduce patient susceptibility to relapse or reinfection. Fecal microbiota transplantation (FMT) in particular has garnered significant clinical interest. FMT transfers fecal microbiota from a healthy individual to a CDI patient to restore a healthy gut microbiota.

Prevention

Not all people who acquire *C. difficile* develop CDI; thus prevention measures can target reducing both the spread of the bacteria or spores and patient susceptibility to infection. One study statistically modeled CDI within the hospital setting and suggested that reducing patient susceptibility to infection is more effective in reducing CDI cases than lowering transmission rates.[20] The likelihood of developing CDI depends on a number of factors that allow colonization and toxin production, including failure of the immune defenses and use of antibiotics, particularly broad-spectrum or multiple antibiotics. Known risk factors for CDI include older age, comorbidities, and use of gastric acid suppressant medications.[21] Mortality is associated with age, white blood cell count, serum albumin, and serum creatinine.[21] Risk profiles for recurrent CDI are similar.[22] Recent prevention efforts have included eliminating, where possible, the offending antibiotic, and using environmental and infection control strategies, as well as seeking to improve immune defenses through healthy digestive function and gut flora and improved nutrition.[23]

Preventing the spread of *C. difficile* within institutional settings depends on staff compliance with national guidelines and standards[24] and locally determined hygiene protocols. Unfortunately, protocols for some targeted hospital-acquired infections may not be effective against *C. difficile*. For example, the availability of alcohol hand rubs improved physician

compliance and reduced Methicillin-resistant staphylococcus aureus (MRSA) infections,[25] yet *C. difficile* produces spores that can withstand hostile environments and are resistant to alcohol hand rubs and other routine antiseptics. Spores may be best removed by handwashing. Other institutional prevention strategies may be required as *C. difficile* transmission knowledge develops. For example, one study isolated *C. difficile* spores and cultured the bacterium from air samples in a United Kingdom hospital 4 to 7 weeks after the last confirmed CDI case in the ward.[26]

Scope and Key Questions

Scope of the Review

In December 2011, the Agency for Healthcare Research and Quality (AHRQ) published the results of Comparative Effectiveness Review (CER) No. 3, Effectiveness of Early Diagnosis, Prevention, and Treatment of *Clostridium difficile* Infection, prepared by the Minnesota Evidence based Practice Center.[15] This CER examined the evidence on the sensitivity and specificity of *C. difficile* infection laboratory diagnostic tests, the effectiveness of prevention strategies, and the effectiveness and harms of antibiotic and adjuvant treatments for adults with CDI. In January 2014, AHRQ published a surveillance report assessing whether an update of CER No. 3 was warranted. The report found new evidence for all key questions, suggesting the results were out of date.[27]

Several main findings were reported in CER No. 3. For diagnostic testing, direct comparisons of commercially available enzyme immunoassays for *C. difficile* toxins A and B found no major differences in sensitivity or specificity. Limited evidence suggested that tests for genes related to *C. difficile* toxins production may be more sensitive than immunoassays, but that specificities were inconsistent. Moderate-strength evidence in favor of antibiotic restriction policies for prevention was found. While no antimicrobial was clearly superior for the initial cure of CDI, as noted above, recurrence was less frequent with fidaxomicin than with vancomycin. Many potential new treatments were examined, and of these, fecal microbiota transplants for multiple recurrences appeared promising. However, with the numerous new publications identified in the surveillance report, an update of the review was merited.

This update systematically reviewed and assessed the evidence for diagnosis, prevention, and treatment of *C. difficile* using the original report and newly available evidence. We used essentially the same search strategy and review methodology, minimally updated to meet current review methods guidance. We made some minor modifications to the key questions in order to focus the update on current clinical concerns and due to the scarce literature base. Specifically, we deleted several subquestions regarding treatment effectiveness for subgroups. Since there has been some growth in the diagnostic testing literature, and diagnostic testing continues to be an area of decisional conflict, we also added a subquestion for testing strategy effects on final patient or health system outcomes.

Key Questions

KQ1: How do different methods for detection of toxigenic *C. difficile* to assist with diagnosis of CDI compare in their sensitivity, specificity, and predictive values?
   a. Overall?
   b. Do performance measures vary with sample characteristics?

c. Does testing strategy impact patient health or health system outcomes?

KQ2: What are effective prevention strategies?
   a. What is the effectiveness of current prevention strategies?
   b. What are the harms associated with prevention strategies?
   c. How sustainable are prevention practices in health care (outpatient, hospital inpatient, extended care) and community settings?

KQ3: What is the comparative effectiveness and harms of different antibiotic treatments?
   a. Does effectiveness vary by disease severity?

KQ4: What are the effectiveness and harms of nonantibiotic adjunctive interventions?
   a. Overall
   b. In patients with relapse/recurrent CDI.

PICOTS

Table 1 provides the PICOTs by the key questions. The analytic frameworks can be found in Appendix A.

Table 1. Review PICOTS

PICOT	KQ1 Included	KQ1 Excluded	KQ2 Included	KQ2 Excluded	KQ3-4 Included	KQ3-4 Excluded
Population	• Adults with clinical signs consistent with CDI	• Pediatric patients alone • Patients not suspected to have CDI; healthy subjects • Patients already diagnosed with CDI	• Primary prevention: Adults at risk for CDI • Recurrence prevention: Adults with clinical signs consistent with CDI	• Pediatric patients	• Adults with clinical signs consistent with CDI • Adjunctive to prevent CDI: Adults at risk for CDI • Adjunctive to prevent recurrence: Adults with clinical signs consistent with CDI	• Pediatric, nonhuman, in vivo, or healthy volunteers.
Intervention	• Diagnostic tests for toxin producing *C. difficile*: • Immuoassays (enzyme immunoassays [EIA], enzyme-linked immunoassays [ELISA], immunochromato-graphy assays) • Tests for toxins • Two step strategies • DNA amplification (polymerase chain reaction [PCR], loop-mediated isothermal amplification [LAMP])	• Tests of stool culture alone. • Tests to validate a technique in "known" or proven samples. • Tests in which the reference standard is not applied to all samples. • Tests examining cost characteristics. • Tests not commercially available in the U.S. • Tests only typing *C. difficile* strains. • Tests establishing proof of concept for new testing techniques (such as fecal calprotectin)	• Antibiotic stewardship, education, bundled preventive programs, prebiotics or probiotics used as preventive measures • Hospital inpatient environmental cleaning, monitoring, or surveillance • Environmental cleaning for long-term care facilities.	• None	• Standard antibiotic treatments: o Metronidazole o Rifaxamin o Vancomycin o Fidaxomicin • Nonantibiotic adjunctive treatments: o Fecal transplant o Immunoglobulin o Pre/probiotics o Toxin binding agents o Rifampicin • Other new treatments available in the U.S.	• Treatments approved outside of the U.S. that are not available in the U.S.
Comparator groups	• Reference Standard: cell cytotoxicity assay and/or toxigenic stool culture • Comparators: any includable diagnostic	• In-house laboratory tests not commercially available.	• Usual prevention practices for prevention strategies	• None	• Standard antibiotic treatments: active treatments such as metronidazole or vancomycin. • Nonantibiotic adjunctive	• None

PICOT	KQ1 Included	KQ1 Excluded	KQ2 Included	KQ2 Excluded	KQ3-4 Included	KQ3-4 Excluded
	test listed above as intervention. • For health system and patient outcomes: historical data comparators may be used.				treatments: placebo, active controls, usual care.	
Outcomes	• Sensitivity • Specificity • Predictive values • Time-to-results • Patient outcomes • Health system outcomes	• None	• CDI incidence rates • CDI complication rates • CDI mortality rates • Harms, such as increase in organism resistance, hospital cleaning staff safety (bundled prevention programs), infection by introduced probiotics, isolation harms. • Intermediate Outcomes o Appropriate antibiotic use o Positive environmental cultures. o Days to resolution of symptoms (shorter window for transmission). • Other prevention strategy-related process variable demonstrating prevention strategy was taken up.	• Studies that do not report CDI incidence rates and tie incidence to the inter-mediate process measures	• Mortality • Recurrence (study author defined) • Clearance (study author defined) • Complications • CDI-related colectomy rate • Symptom resolution (study author defined) • Harms, such as delayed treatment response	• None
Timing	• Time to test results • For patient or health system outcomes: no specific time requirement	• None	• Variable	• None	• Variable, generally from 4 weeks to several months	• None
Setting	• Healthcare facilities: outpatient, inpatient, extended	• None	• Healthcare facilities: outpatient, inpatient, extended care.	• None	• Healthcare facilities: outpatient, inpatient, extended care	• None

CDI=*Clostridium difficile* infection; EIA=enzyme immunoassay; ELISA=enzyme-linked immunoassay LAMP=loop mediated isothermal amplification; PCR=polymerase chain reaction

Methods

The methods for this comparative effectiveness review (CER) update follow the methods suggested in the AHRQ Methods Guide for Effectiveness and Comparative Effectiveness Reviews (available at http://www.effectivehealthcare.ahrq.gov/methodsguide.cfm); certain methods map to the PRISMA checklist.[28] All methods and analyses were determined *a priori*. We recruited a technical expert panel to provide high-level content and methodological expertise feedback on the review protocol. This section summarizes the methods used.

Literature Search Strategy

Our search methods were essentially the same as were used for CER No. 3. We searched Ovid MEDLINE, and Cochrane Central Register of Controlled Trials (CENTRAL) from 2011 to the present to update CER No. 3. The keyword search for 'difficile' is highly specific yet sensitive to *C. difficile* related articles. The search algorithm is provided in Appendix B.

We conducted additional grey literature searching to identify relevant completed and ongoing studies. Relevant grey literature resources included trial registries and funded research databases. We searched ClinicalTrials.gov and the International Controlled Trials Registry Platform (ICTRP) for ongoing studies. Scientific information packet (SIP) letters and emails were sent to relevant industry stakeholders to request submission of published and unpublished information on their product(s). Grey literature search results were used to identify studies, outcomes, and analyses not reported in the published literature to assess publication and reporting bias and inform future research needs.

We will update searches while the draft report is under public/peer review.

Studies were included in the review based on the PICOTS framework outlined in Table 1 and the study-specific inclusion criteria described in Table 2.

Table 2. Study inclusion criteria

Category	Criteria for Inclusion
Study Enrollment	Studies that enroll adults with suspected CDI
Study Design and Quality	Any: Systematic reviews with relevant questions of fair or good quality (see Risk of Bias section below); must include risk of bias assessment with validated tools. Diagnosis: Studies of diagnostic accuracy assessing the operating characteristics of commercially available diagnostic test(s) for CDI in adult patients suspected of having CDI that include CCNA or toxigenic culture as the reference standard applied to all samples. Prevention: RCTs, nonrandomized controlled trials, prospective cohort studies, retrospective cohort, time series, and before/after trials will be included. Cohort studies must include a comparator and appropriate methods to correct for selection bias. Standard Treatment: RCTs, nonrandomized controlled trials, and prospective cohort studies will be included for each population and treatment option. Prospective studies must include a comparator and appropriate methods to correct for selection bias. Studies specifically addressing treatment harms may also include retrospective and case series designs. Nonantibiotic standard treatment: RCTs, nonrandomized controlled trials, prospective cohort studies, and case series (at least 10 subjects) will be included for each population and treatment option. Prospective studies must include a comparator and appropriate methods to correct for selection bias. Studies specifically addressing treatment harms may also include retrospective and case series designs. For all KQ: Observational studies that do not adequately report study information to allow the abstraction of time sequences for treatment and followup duration or have

Category	Criteria for Inclusion
	indeterminable numerators and denominators for outcomes and adverse event rates were excluded at the abstraction phase.
Time of Publication	Update from previous systematic review. We scanned 2010 forward to assure all published literature was identified.
Publication Type	Published in peer reviewed journals
Language of Publication	English language publications will be included because that literature best represents interventions available in the United States. However, the search was not limited by language so that potential language bias could be assessed

CCNA-cell cytotoxicity neutralization assay; CDI=*Clostridium difficile* infection; PCR=polymerase chain reaction; RCT-randomized controlled trial

Study Selection and Data Extraction

We reviewed bibliographic database search results for studies relevant to our PICOTS framework and study-specific criteria. All studies identified at title and abstract as relevant by either of two independent investigator underwent full-text screening. Two investigators independently performed full-text screening to determine if inclusion criteria were met. Differences in screening decisions were resolved by consultation between investigators, and, if necessary, consultation with a third investigator. Appendix C provides a list of articles excluded at full text.

We first assessed the relevance of systematic reviews that met inclusion criteria. If we determined that certain key questions or comparisons addressed in the previous systematic review were relevant to our review, we assessed the quality of the methodology using modified AMSTAR criteria.[29] When prior systematic reviews were assessed as sufficient quality, and when the review assessed strength of evidence or provided sufficient information for it to be assessed, we used the conclusions from that review to replace the *de novo* process. If additional studies on these comparisons were identified, we updated the systematic review results. We then abstracted data from eligible trials and prospective cohort studies not included in previous systematic reviews that addressed comparisons not sufficiently addressed by a previous eligible systematic review. One investigator abstracted the relevant information directly into evidence tables. A second investigator reviewed evidence tables and verified them for accuracy.

Risk of Bias Assessment of Individual Studies

Risk of bias of eligible studies was assessed by two independent investigators using instruments specific to each study design. For diagnostic studies, we used the QUADAS-2 tool.[30] For RCTs, questionnaires developed from the Cochrane Risk of Bias tool were used. We developed an instrument for assessing risk of bias for observational studies based on the RTI Observational Studies Risk of Bias and Precision Item Bank[31] (Appendix D). We selected items most relevant in assessing risk of bias for this topic, including participant selection, attrition, ascertainment, and appropriateness of analytic methods. Study power was assessed in 'other sources of bias' in studies with data that were not eligible for pooling. Overall summary risk of bias assessments for each study were classified as low, moderate, or high based upon the collective risk of bias inherent in each domain and confidence that the results were believable given the study's limitations. When the two investigators disagreed, a third party was consulted to reconcile the summary judgment.

Data Synthesis

Evidence and summary tables followed those used for CER No. 3 wherever possible. Information from individual studies reviewed in CER No. 3 were brought forward into this updated report when meta-analysis was performed using such information. Otherwise, tables show studies identified for the update and text notes if and how overall results from CER No. 3 were amended.

Where possible, we used data from previous reviews combined with data abstracted from newly identified studies to create new datasets for analysis. We summarized included study characteristics and outcomes in evidence tables. We emphasized patient-centered outcomes in the evidence synthesis. We used statistical differences to assess efficacy and comparative effectiveness and calculate the minimum detectable difference that the data allowed (β=.8, α=.05).

For diagnostic studies we looked at the reference standards and base contrasts on the type of reference standard and respective operating characteristics.[32,33] We focused on the differences between test category/methodology sensitivities and specificities rather than on specific test sensitivities and specificities themselves. Categories were Immunoassays for Toxin A/B, glutamate dehydrogenase (GDH), PRC, LAMP, and test algorithms. We pooled one-step NAAT (PCR or LAMP) studies using random effects models; diagnostic test algorithm studies that include NAAT tests (likely PCR) were pooled with other test algorithms. Data were analyzed in OpenMetaAnalyst. We calculated sensitivity, specificity, receiver operating characteristic curves (ROC), and negative and positive likelihood ratios.[34] We used random effect models to pool data when clinically appropriate.

For studies that used multiple reference standards, such as culture, toxigenic culture, and CCNA, we used toxigenic culture as the reference standard. If different reference standards were used for specific subgroups (such as study site) and none was used across all the samples, then we used the reference standard that was used in interpretation of the index test.

For treatment studies, if certain comparisons could be pooled, we conducted meta-analyses using a random effects model. Data were analyzed in Stata I/C version 12.1.We calculated risk ratios (RR) and absolute risk differences (RD) with the corresponding 95 percent CI for binary primary outcomes. Weighted mean differences (WMD) and/or standardized mean differences (SMD) with the corresponding 95 percent confidence intervals (CIs) were calculated for continuous outcomes. We assessed the clinical and methodological heterogeneity and variation in effect size to determine appropriateness of pooling data.[35] We assessed statistical heterogeneity with Cochran's Q test and measure magnitude with I^2 statistic.

Strength of Evidence for Major Comparisons and Outcomes

The overall strength of evidence for select outcomes within each comparison were evaluated based on four required domains: 1) study limitations (internal validity); 2) directness (single, direct link between intervention and outcome); 3) consistency (similarity of effect direction and size); and 4) precision (degree of certainty around an estimate).[36] A fifth domain, reporting bias, was assessed when strength of evidence based upon the first four domains was moderate or high.[36] Based on study design and conduct, risk of bias was rated as low, medium, or high. Consistency was rated as consistent, inconsistent, or unknown/not applicable (e.g., single study). Directness was rated as either direct or indirect. Precision was rated as precise or imprecise. Other factors that may be considered in assessing strength of evidence include dose-response

relationship, the presence of confounders, and strength of association. Based on these factors, the overall evidence for each outcome was rated as:[36]

- **High:** Very confident that estimate of effect lies close to true effect. Few or no deficiencies in body of evidence, findings believed to be stable.
- **Moderate:** Moderately confident that estimate of effect lies close to true effect. Some deficiencies in body of evidence; findings likely to be stable, but some doubt.
- **Low:** Limited confidence that estimate of effect lies close to true effect; major or numerous deficiencies in body of evidence. Additional evidence necessary before concluding that findings are stable or that estimate of effect is close to true effect.
- **Insufficient:** No evidence, unable to estimate an effect, or no confidence in estimate of effect. No evidence is available or the body of evidence precludes judgment.

Applicability

Applicability of studies was determined according to the PICOTS framework. Study characteristics that may affect applicability include, but are not limited to, the population from which the study participants are enrolled, diagnostic assessment processes, narrow eligibility criteria, and patient and intervention characteristics different from those described by population studies of *C. difficile*.[37]

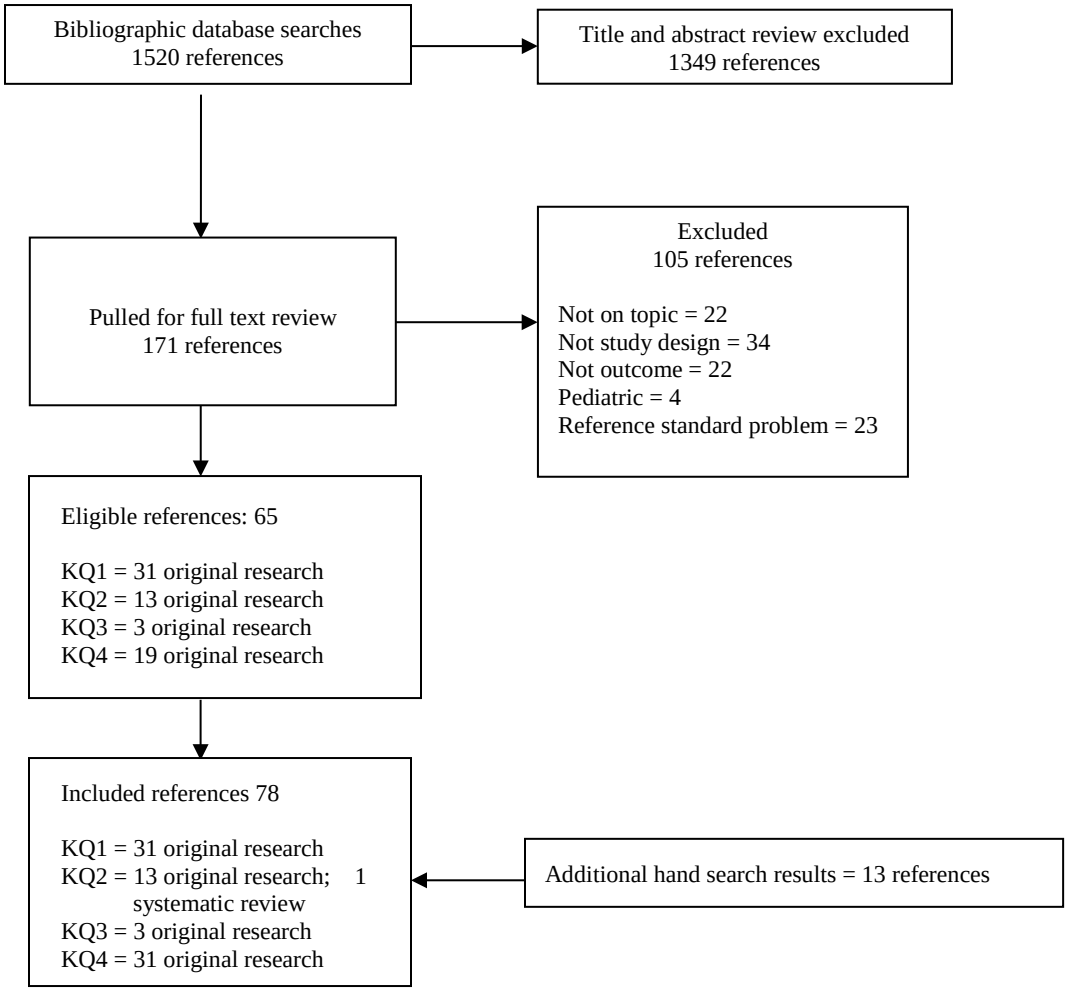
Applicability of studies of diagnostic accuracy of diagnostic tests for CDI may be influenced by the selection of patient samples in the studies included and the degree (if any) of delineation of the demographic and clinical characteristics of the studies' respective patient populations and how these characteristics compare with a local population. Further, certain diagnostic tests may not be available to all clinicians depending on local health system factors.

Results

Literature Search Results

We identified 1520 unique citations (Figure 1) from 2010 to August 2014. After excluding articles at title and abstract, full texts of 171 articles were reviewed to determine final inclusion. Thirteen articles were added through hand search.

Figure 1. Literature flow diagram



The appendices of this report provide detailed information about the included studies: evidence tables (Appendix E); risk of bias and quality assessments of original research and systematic reviews (Appendix F); detailed analyses (Appendix G); and detailed strength of evidence assessments (Appendix H).

Key Question 1. How do different methods for detection of toxigenic *C. difficile* to assist with diagnosis of CDI compare in their sensitivity, specificity, and predictive values?

Thirty-one new studies evaluated diagnostic tests for CDI. Nineteen studies were from Europe, five from the United States, two from Canada, three from Korea, and one each from Mexico and Saudi Arabia. Twenty-three studies were performed at a single center and eight studies were multicenter studies. (See Appendix C for evidence tables.) Overall, these studies, when combined with the 13 studies from the original review, include data on eight named immunoassays for *Clostridium difficile* toxins A and B, four GDH tests, eight test algorithms, one LAMP, and nine PCR. The number of studies assessing the diagnostic accuracy of tests that detect genetic material from *Clostridium difficile* in feces (LAMP and PCR) increased considerably − 14 studies in the update compared to only three in the original review.

Table 3 provides a summary of the findings.

Table 3. Summary of diagnostic test findings new with the update

Diagnostic Test	Study Information	Findings	Strength of Evidence
NAAT (LAMP and PCR) Tests	11 LAMP arms (1 test type), 25 PCR arms (9 test)	Sensitive (LAMP 0.95, CI 0.89 - 0.97; PCR 0.94, CI 0.92 - 0.96) and specific (LAMP 0.98, CI 0.96 -0.99; PCR 0.97, CI 0.96-0.98) for CDI	High (low study limitation, consistent, precise)
Tests for Toxin A/B	57 arms (8 test types)	Insensitive (0.71, CI 0.67 - 0.75) but specific (0.98, CI 0.97- 0.99) for CDI	Moderate (low study limitation, consistent, imprecise)
Tests for GDH	7 arms (4 test types)	Sensitive (0.94, CI 0.89 – 0.97) but less specific (0.94, CI 0.89 – 0.97) for CDI	Moderate (moderate study limitation, unknown consistency, precise)
Test Algorithms	8 arms (8 test types)	Insensitive (0.66, 0.52-0.78) but specific (1.00, 0.99-1.0) tests for CDI	Low (moderate study limitation, consistent, imprecise)

CDI=*Clostridium difficile* infection; GDH=glutamate dehydrogenase; LAMP=loop-mediated isothermal amplification; NAAT=nucleic acid amplification tests; PCR=polymerase chain reaction

The general rankings provided in Table 3 are based on the overall pattern of results summarized in Table 4, which shows the sensitivity, specificity, and negative and positive likelihood ratios (forest plots and ROCs in Appendix G) comparisons. In short:

- A positive LAMP assay is likely more effective at increasing the probability that a patient has CDI than PCR, Toxin A and/or B tests, and GDH assays but is less effective than algorithmic approaches. A negative LAMP assay is as effective at decreasing the probability that a patient has CDI as PCR and GDH assays and is more effective than Toxin A/B and algorithmic approaches.
- A positive PCR for CDI is more effective at increasing the post-test probability that a patient has CDI than a positive GDH test, similarly effective to LAMP and Toxin A/B assays, and less effective than algorithmic approaches. A negative PCR test is as effective at decreasing the probability that a patient has CDI as LAMP and GDH assays and more effective than Toxin A/B and algorithmic approaches.
- A positive immunoassay for Toxin A and/or B is more effective at increasing the post-test probability that a patient has CDI than a positive GDH test but is less effective at decreasing the probability that a patient has CDI than PCR, LAMP, and algorithmic

approaches. A negative immunoassay for Toxin A and/or B is more effective than algorithmic approaches but is less effective than PCR, LAMP, and GDH tests.

- A positive GDH assay is less effective at increasing the probability that a patient has CDI than all the other test classes but a negative test is as effective at decreasing the probability of CDI (albeit with less precision in the estimate) as PCR and LAMP and more effective than Toxin A and/or B tests and algorithmic approaches.
- A positive test for CDI via an algorithmic test is the most effective approach to increase the post-test probability that a patient has CDI and is the least effective, when negative, at decreasing the probability that a patient has CDI.

Heterogeneity within the classes is not easily explained by test type. The reasons for the differences in the operating characteristics between individual tests within the same class and between classes of tests are not well described in the studies, and while studies were selected to have good internal validity, studies may have differed significantly in the conduct of the tests and the patient populations.

We found no studies that met the inclusion criteria that provided sufficient sample characteristics to evaluate whether performance measures varied systematically based on health system, laboratory, training methods, or patient characteristics. Similarly, no studies evaluated the effect of different assays for CDI on health systems or patient outcomes.

Table 4. Summary of pooled diagnostic tests by test class

Test Characteristics	LAMP	PCR	Toxin A/B	GDH	Test Algorithms
Studies (k)	11	25	57	7	8
Sensitivity	0.95	0.94	0.71	0.94	0.66
95% CI	0.89-0.97	0.92-0.96	0.67-0.75	0.89-0.97	0.52-0.78
I2 for Heterogeneity	78.6	36.55	89.54	66.32	97.77
Specificity	0.98	0.97	0.98	0.94	0.995
95% CI	0.96-0.99	0.96-0.98	0.97-0.99	0.89-0.97	0.99-0.998
I2 for Heterogeneity	92.76	76.7	89.64	96.45	95.32
Positive Likelihood Ratio	57.29	35.45	33.23	15.43	109.65
95% CI	23.09-142.14	25.65-49	25.75-42.88	8.87-26.82	61.2-196.46
I2 for Heterogeneity	92.26	76.42	86.09	96.45	89.12
Negative Likelihood Ratio	0.039	0.037	0.213	0.05	0.29
95% CI	0.021-0.074	0.023-0.058	0.162-0.280	0.025-0.099	0.11-0.74
I2 for Heterogeneity	82.72	89.29	88.03	97.82	96.54

CI= confidence interval; GDH= glutamate dehydrogenase; LAMP= loop-mediated isothermal amplification; NAAT= nucleic acid amplification tests; PCR=polymerase chain reaction

Key Question 2. What are effective prevention strategies?

Seventeen articles examined prevention studies; one systematic review (which included five new studies), one on chlorhexidine gluconate bathing of patients, two on using hydrogen peroxide vapor for room disinfection, two on hand hygiene, one on gloving, and four on multicomponent interventions. None were controlled trials. Two used quasi-experimental designs, four (plus all relevant studies in the systematic review) used interrupted time series analysis, four used prospective pre/post designs of single sites, and one used a retrospective pre/post designs for a single site.

Overall, while study design and reporting improved somewhat from the original review, the evidence available to link prevention strategies to clinically important outcomes, such as CDI incidence, remains low strength. Table 5 provides a summary of the findings.

Table 5. Summary of prevention findings new with the update

Intervention	Study Information	Findings	Strength of Evidence
Antibiotic stewardship	1 systematic review (6 studies)	Appropriate prescribing practices associated with decreased CDI	Low (moderate to high study limitation, consistent, imprecise)
Bathing patients with chlorhexidine gluconate	1 study	Reduced CDI (model estimated RR 0.71, CI 0.57 – 0.89; 3 times per week for all cohorts)	Low (moderate to high study limitation, unknown consistency, imprecise)
Hydrogen peroxide vapor with terminal room cleaning	3 studies		Insufficient (high study limitation, consistent, imprecise)
Pulsed xenon ultraviolet light after terminal room cleaning	1 study		Insufficient (high study limitation, consistent, imprecise)
Handwashing campaigns	1 study	Reduced CDI (rates fell from 16.75 to 9.49 cases per 10,000 bed days)	Low (moderate study limitation, unknown consistency, imprecise)
Multicomponent prevention interventions	4 studies	 Sustainable over several years	Insufficient for effectiveness Low (moderate to high study limitation, consistent, imprecise)

CDI=*Clostridium difficile* infection; CI= confidence interval; RR=relative risk

Antibiotic Stewardship

One new high-quality systematic review[38] of antibiotic stewardship practices in inpatient settings included six studies (one RCT and five interrupted time series) and overlapped with the original review by one interrupted time series study.[39] The new systematic review categorized stewardship practices into audit and feedback, formulary restrictions and preauthorization interventions, guidelines implemented with feedback, guidelines without feedback, and computerized decision support programs. The six studies were evaluated as providing low strength of evidence that antibiotic stewardship programs reduced CDI incidence within the four antibiotic use program categories examined (audit and feedback, formulary restrictions, guidelines with feedback, and computerized decision support).[38] The review found no reports of harms associated with stewardship programs.

Transmission Interruption

Bathing patients was a new form of transmission interruption from the updated literature. One moderate risk of bias study found chlorhexidine gluconate bathing either 3 days per week or daily in three cohorts within one hospital reduced CDI rates.[40] While there were no concurrent controls, the cohort design allowed for some replicability and comparison. Changes in effects with dosing (daily vs. three times per week) and the wash-out period strengthen the finding. Highest compliance rates were found in the ICU cohort versus general hospital or medical/surgical cohorts; however, regression models did not find compliance associated with CDI rates; the model estimated RR 0.71 (CI 0.57 – 0.89) three times per week for all cohorts.

Two new studies examined terminal room cleaning including hydrogen peroxide vapor.[41,42] Rooms known to have prior occupants with CDI or other disease-causing organisms are sealed and sporicidal hydrogen peroxide vapor is released into the rooms in a gassing process. Protocols are followed for sealing the vapors within the rooms until proper ventilation is complete. One

pre/post study in the original review used hydrogen peroxide vapor as part of a multicomponent intervention to respond to an abrupt increase in nosocomial CDI infections (Table 4 in the original report). The decrease in CDI infections could not be separated from the natural decline that follows epidemics, nor could the effect of the vapor be separated from the multiple component intervention. Both new studies occurred in large (900-bed) hospitals not facing epidemic or hyperendemic events. One pre/post study of the vapor versus standard cleaning with bleach found a statistically significant reduction in CDI incidence.[41] In contrast, the quasi-experimental cohort study used hydrogen peroxide liquid in the standard cleaning solution and found a trend in reduction but no statistical difference in CDI.[42] Of the three studies, cleaning time ranged from 2 hours 20 minutes to 3 – 4 hours per room.

One new pre/post study examined the effect of portable pulsed xenon ultraviolet light after terminal room cleaning on CDI incidence in a single 140-bed community hospital.[43] Rooms with a previous CDI patient were cleaned with a chlorine-based disinfectant product, followed by one 7-minute exposure in the bathroom and two 7-minute exposures in the main room to the ultraviolet light. The lights were also used in the operating suites at night, emergency departments in the early mornings, and other clinical areas as available. CDI incidence and hospital-acquired CDI associated deaths and colectomies were found to decline. However, in addition to not including a control group, the hospital also added ciprofloxacin to the pharmacy formulary, with a resulting decline in levofloxacin and quinolone.

Two new studies examined the effect of handwashing campaigns on CDI rates using uncontrolled interrupted time series design in 187 hospital trusts in the England[44] and 166 acute care hospitals in Ontario, Canada.[45] The original report did not locate studies that directly addressed the effect of handwashing on CDI. The two new studies examined campaigns that incorporated education and training programs and monitoring and feedback through either internal reports[44] or public reporting.[45] The program in England also empowered patients to remind healthcare workers of hand hygiene.[45] Based on the one moderate risk of bias study from England, low-strength evidence suggests that handwashing campaigns can reduce CDI incidence over a 3-year period, with rates falling from 16.75 to 9.49 cases per 10,000 bed days. The study also found via regression model that soap use (measured via centralized procurement of soap) was independently associated with a slight reduction in CDI.[44] The other high risk of bias study found no statistical difference; however, the authors note having been unable to adjust for several possible confounders, including patient location in the hospital, type of hand product used, and concomitant introduction of other hospital-level infection prevention and control interventions.

One new pre/post study examined universal gloving with emollient-impregnated gloves.[46] This study does not add significantly to the original report's finding of low-strength evidence for gloving based on one RCT. (Table 4 in the original report)

Cleaning and disinfection studies reported no adverse events noted for chlorhexidine gluconate bathing[40] or hydrogen peroxide vapor.[41,42] Harms were not reported for antimicrobial stewardship programs.

Multiple Component Studies

Four new high-risk-of-bias studies used multiple component interventions to address reducing CDI rates.[47-50] Two used pre/post designs and two used uncontrolled interrupted time series approaches (both at single hospitals). Ten studies with pre/post or time series with pre/post statistical approaches were identified for the original review (Table 4 in the original report).

Some differences in the literature from the original review are noted. First, the studies were framed as responding to general heightened concerns for CDI as a hospital acquired pathogen, rather than a localized epidemic or high endemic. Second, study followup was longer, ranging from 2 – 3 years for pre/post studies[47,48] to 27 – 81 months for time series studies.[49,50] Third, studies tended to include more information on CDI definitions and laboratory testing methods.

The study designs do not permit inferences for individual intervention components; however, the increase in study periods suggests that multiple component interventions can be sustained over several years.

Key Question 3: What is the comparative effectiveness and harms of different antibiotic treatments?

Three studies met inclusion criteria: an RCT comparing fidaxomicin to vancomycin,[51] a three-arm RCT comparing tolevamer (a toxin-binding resin) to metronidazole and vancomycin,[52] and a three-arm prospective cohort study comparing intravenous metronidazole to oral metronidazole and vancomycin.[53] Data from these new studies were combined with studies from the original report—a previous RCT of fidaxomicin versus vancomycin, and with three previous RCTs comparing metronidazole and vancomycin—to assess the efficacy of each drug.

Table 6 provides a summary of the findings.

Table 6. Summary of standard treatment findings using pooled RCT data from original report and update

Intervention	Study Information	Findings	Strength of Evidence
Vancomycin vs. Metronidazole	4 RCTs N=872	Initial Cure: favors vancomycin 83.9% vs. 75.7%; RR 1.08, 95% CI 1.02 – 1.15	High (moderate study limitation, consistent, precise)
	N=705	Recurrent CDI: not significantly different 16.5% vs. 18.7%; RR 0.89, 95% CI 0.65 – 1.23	Moderate (moderate study limitation, imprecise, consistent)
Fidaxomicin vs. Vancomycin	2 RCTs N=1,111	Initial Cure: not significantly different 87.6% vs. 85.6%; RR 1.02, 95% CI 0.98-1.07	Moderate (low study limitation, consistent, imprecise)
	N=962	Recurrent CDI: favors fidaxomicin 14.1% vs. 26.1% RR 0.55, 95% CI 0.42-0.71	High (low study limitation, consistent, precise)
Any intervention: Treatment effect by disease severity	3 RCTs	Treatment results did not differ by disease severity	Low (moderate to high study limitation, inconsistent, imprecise)

CDI=*Clostridium difficile* infection; CI= confidence interval; RR=relative risk

Benefits

The findings that vancomycin is more effective for initial cure of CDI in adults is new to this update because of improved precision. While the results for fidaxomicin versus vancomycin are consistent with the original review, the strength of the evidence improved.

An observational study (n = 205) comparing oral metronidazole, intravenous metronidazole, and vancomycin was also identified.[53] Results are similar to the RCTs, so this study was not included in the analyzed set. Initial cure was comparable for oral vancomycin (81.0%) and oral

metronidazole (82.6%), but was significantly lower for intravenous metronidazole (52.4%; P <.001). Intravenous metronidazole performed significantly worse than either oral drug.

Time to resolution of diarrhea was reported in both the newly identified RCTs, with no differences observed based on treatment received. This outcome was not reported in the observational study. For both time to resolution of diarrhea and mortality, results did not differ from the original review's finding of no differences.

Harms

Only a slight change was observed based on the newly included studies. Similar to the original report, in the trial of metronidazole versus vancomycin, a similar percentage of subjects in each treatment arm experienced one or more serious adverse event. However, more subjects in the metronidazole group discontinued study medication because of an adverse event (11.2% vs. 6.5%; P = .06), whereas more subjects in the vancomycin group had evidence of nephrotoxicity (4.6% vs. 1.0%, P = .02).

Disease Severity

In both new RCTs, pre-specified subgroup analyses among subjects with severe disease were performed to assess differences in outcome by treatment arm. Disease severity was generally determined by one or more clinical values such as white blood cell counts, serum creatinine concentrations, body temperature, and severity of abdominal pain due to CDI. No significant differences were observed for initial cure for severity subgroups. Analyzing by disease severity did not change the overall study results. One study found less recurrence for vancomycin versus metronidazole for severe disease, but the results varied based on whether per-protocol, modified intention to treat, or strict intention to treat analyses were used. The observational study also looked for a treatment effect when stratified by disease severity and found no significant differences. The original review found insufficient evidence for treatment by severity based on one *post hoc* subgroup analysis for vancomycin versus metronidazole.

Key Question 4: What are the effectiveness and harms of nonantibiotic adjunctive interventions?

Nonadjunctive treatments were categorized as fecal microbiota transplant (FMT), probiotics, or other. FMT was the largest updated literature set for non-antibiotic adjunctive therapy. Nineteen new studies examined FMT for CDI: two RCTs and 17 observational studies, in addition to two observational studies carried forward from the original review. We identified 10 new studies on probiotic use: nine RCTs and one observational study, in addition to seven RCTs included in the prior report. We identified two new RCTs on other nonstandard therapies.

Table 7 summarizes the findings.

Table 7. Summary of nonstandard treatment findings using data from original report and update

Intervention	Study Information	Findings	Strength of Evidence
FMT	2 RCTs, 19 case series N=516	(Unpooled) Resolves diarrhea and prevents relapse in patients with recurrent CDI	Low (high study limitation, consistent, precise)
	2 contributing case series N=5	Refractory CDI	Insufficient (high study limitation, imprecise, unknown)
Lactobacillus vs. placebo	6 RCTs N=584	Prevent CDI: favors lactobacillus RR 0.27, 95% CI 0.15-0.49	Low (moderate to high study limitation, consistent, imprecise)
S. boulardii vs. placebo	6 RCTs N=588	Prevent CDI: not significant RR 0.77, 95% CI 0.38-1.54	Low (high study limitation, consistent, imprecise)
Multiorganism probiotics vs. placebo	4 RCT N=1723	Prevent CDI: not significant RR 0.48, 95%, CI 0.19-1.21	Low (high study limitation, consistent, imprecise)

CDI=*Clostridium difficile* infection; CI= confidence interval; RCTs=randomized controlled trials; RR=relative risk

FMT for Recurrent CDI

Nineteen new studies addressed FMT for recurrent CDI; two were small size RCTs and the others were case series. Most studies were small, enrolling 12 to 70 individuals. Followup was variable, and ranged from 3 weeks to 8 years.

The two RCTs are noteworthy. One unblinded, three-arm RCT, conducted in the Netherlands, enrolled 43 adults with recurrent CDI (mean age 70, 43 percent women).[54] Patients were randomized to oral vancomycin, FMT, or vancomycin plus bowel lavage. Followup was 10 weeks and the endpoint was resolution of diarrhea. The study was stopped early due to a large difference between the FMT and comparator groups (81% vs. 31% and 23%), largely due to an unexpectedly low response rate in the group randomized to vancomycin. FMT was administered via nasoduodenal tube. The resolution of diarrhea rate in the two vancomycin arms was considerably lower than the anticipated 60 percent. This may have been due to chance, and the 60-percent rate may have been achieved had the study treated the expected 38 patients per arm. However, without having run the full course, the study effect size remains uncertain.

Youngster and colleagues conducted an unblinded RCT that randomized 20 individuals with recurrent CDI (mean age 54) to colonoscopic or nasogastric administration of FMT.[55] The study endpoint was resolution of diarrhea without relapse within 8 weeks. The authors found no difference between the two modalities of FMT administration, with an overall success rate of 70 percent after one treatment.

Based on a qualitative analysis of the unpooled data (Appendix G), low-strength evidence showed that FMT resolves diarrhea and prevents relapse in people with recurrent CDI.

FMT for Refractory CDI

Two studies reported outcomes for FMT in individuals with refractory CDI (defined as an episode that did not respond to antibiotic treatment; clearly identified by study authors). Both were from case series, totaling five individuals.[56,57] Overall, there was insufficient strength of evidence supporting the role of FMT in refractory CDI. Unfortunately, few FMT studies

provided detailed patient information to identify whether included patients could be considered refractory.

Probiotics for CDI

Seventeen studies reported use of probiotics as adjunctive treatment for CDI: nine RCTs and one observational study were newly identified, while seven RCTs were included in the prior report. With 16 RCTs to provide a best evidence base, the observational study will not be discussed further.

In all studies, probiotics were administered as an adjunct to standard antibiotic treatment to prevent CDI. All studies included adult inpatients or outpatients with a mean reported age of 55 to 76 years. The studies enrolled 40 to 2981 subjects. The probiotics tested were lactobacilli species in seven studies, saccharomyces species (*S. boulardii*) in six studies, both lactobacillus and saccharomyces species in one study, lactobacillus and bifidobacterium in two studies, and VSL#3 in one study.

For quantitative analysis, we categorized probiotics as single organism (lactobacillus organisms only), *S.boulardii*, or multiple organism (e.g., multistrain preparation of *lactobacilli* and *bifidobacteria*). Overall, we found low-strength evidence that probiotics containing only lactobacillus organisms are more effective than placebo in preventing an acute episode of CDI, predominantly driven by one moderate risk of bias study that also demonstrated dose response. We found low-strength evidence that probiotics containing S.boulardii given as adjunct to standard antimicrobial therapy are comparable to placebo in preventing an episode of CDI. We also found low-strength evidence that the multiorganisms tested performed no better than placebo.

Other Adjunctive Treatment Agents for CDI

Rifaximin versus placebo after standard antibiotic for CDI was examined by Garey and colleagues.[58] Rifaximin is a nonabsorbable antibiotic with FDA approval to treat traveler's diarrhea. Sixty-eight individuals with CDI (mean age 61, 50 percent were women) were treated for 20 days. After 3 months of followup, authors reported no statistically significant difference in recurrent CDI between groups. Recurrent diarrhea was reported less likely in the rifaximin group, but this included self-reported diarrhea episodes without confirmed *C. difficile* toxins.

Human recombinant lactoferrin versus placebo was examined by Laffan and colleagues.[59] Human recombinant lactoferrin from breast milk has both anti-inflammatory and antimicrobial properties. The study randomized 30 residents of a long-term care facility beginning a new course of antibiotic, either with CDI or without, to human recombinant lactoferrin or placebo for 8 weeks. Mean age was 62, 64 percent were women, and 32 percent were black. The study endpoint was CDI incidence rates at days 14, 42, and 56. CDI rate did not differ statistically between groups.

Harms of Adjunctive Treatments

Harms for FMT were available in the updated literature set. Adverse events after FMT in the single small RCT were diarrhea, cramps, belching and nausea, and constipation.[54] Serious adverse events included one hospitalization and two cases of infections, unrelated to FMT. Upper gastrointestinal bleeding was reported in one study with nasogastric administration of FMT.[60] Other serious adverse events were peritonitis, pneumonia, and microperforation of the

colon.[61] All-cause mortality after FMT ranged from 0 – 25% when reported, depending on the length of followup. Mortality rates after FMT were higher in individuals with refractory compared with recurrent CDI. However, variable followup time, differences in baseline comorbidities, and especially the lack of any control group make placing this figure into context difficult. Whether deaths were due to FMT or reflected the overall poor health status of individuals undergoing FMT was unclear, particularly for those with refractory CDI. While one study reported a followup interval of up to 8 years,[62] the followup for the majority of studies was 3 months or less. Therefore, the long-term (greater than 3 months) adverse effects of FMT are largely unknown.

Fifteen of 17 studies of probiotics as adjunctive treatment for CDI reported data on adverse events (see Appendix Table E7). Treatment with probiotics was not associated with increased risk of adverse events in any of the studies. No serious adverse events were reported that were attributed to probiotic treatment, although followup was typically 4 weeks or less, with two RCTs extending followup to 12 weeks. Given the importance of the potential harm due to probiotics, we reiterate from the original report that fungemia may be a serious potential harm associated with administration of probiotics for CDI in critically ill patients.[63]

Discussion

Overview

This update identified a few notable changes from the original review to support the diagnostic, preventive, and treatment practices for *Clostridium difficile* infection (CDI). Table 8 provides a summary of the findings presented in this update along with the findings of the original report.

Table 8. Summary of findings for update and original review

Key Questions	Level of Evidence Update	Level of Evidence Original Report	Summary/Conclusion/Comments
KQ1 - Diagnostics			
Nucleic acid amplification tests	High	NA	Sensitive and specific for CDI
Enzyme tests for toxins A/B	Moderate	NA	Sensitive but less specific for CDI
Assay tests for glutamate dehydrogenase	Moderate	NA	Specific but less sensitive for CDI
Test Algorithms	Low	NA	Multi-step tests specific but less sensitive for CDI
KQ2 - Prevention			
Antibiotic use	Low	Low	Appropriate prescribing practices associated with decreased CDI
Gloves	Low	Low	Use of gloves in hospital settings reduced CDI incidence
Disposable thermometer	NA	Low	Use of disposable thermometers in hospital settings reduced CDI incidence
Bathing patients, chlorhexidine gluconate	Low	NA	Bathing patients with chlorhexidine gluconate in hospital settings reduced CD incidence
Handwashing/ alcohol gel	Low NA	Insufficient Low	Handwashing campaigns in hospital settings reduce CDI incidence No significant differences in CDI incidence for alcohol gel to reduce MRSA transmission.
Disinfection	NA Insufficient Insufficient	Low Insufficient NA	Intensive disinfection with chemical compounds (hypochlorite, aldehydes, hydrogen peroxide) that kill *C. difficile* spores reduced CDI incidence Hydrogen peroxide vapor treatment with terminal room cleaning evidence remains insufficient Pulsed xenon ultraviolet light after terminal room cleaning evidence insufficient
Multiple component strategies	Insufficient	Insufficient	Body remains insufficient to draw conclusions.
Sustainability	Low	Insufficient	Longer-term studies of prevention program roll-outs suggest programs are sustainable
KQ3 – Standard Treatment			
Vancomycin versus Metronidazole	High Moderate	Moderate Low	Vancomycin more effective in achieving initial cure No difference between groups for recurrent CDI
Fidaxomicin	Moderate	Moderate	No significant differences in initial cure.

Key Questions	Level of Evidence Update	Level of Evidence Original Report	Summary/Conclusion/Comments
versus Vancomycin	High	Moderate	Decreased recurrence among those receiving fidaxomicin
Effect by disease severity	Low	Insufficient	Reported results by treatment arm are present regardless of severity
All other comparisons of standard treatments	NA	Low for all comparisons	Vancomycin versus bacitracin, vancomycin versus nitazoxanide, vancomycin high versus low dose, metronidazole versus nitazoxanide, and metronidazole versus metronidazole plus rifampin. No differences
Strain of organism	NA	Low	One RCT (fidaxomicin versus vancomycin) demonstrated decreased recurrence among those receiving fidaxomicin when the infecting organism was a non-NAP1 strain
Patient characteristics	NA	Insufficient	No comparative data were available
Resistance of other pathogens	NA	Insufficient	No data were available
KQ4 – Adjunctive Treatment			
Treating CDI, active control	NA	Low	Probiotics, prebiotics, *C. difficile* immune whey, and colestipol, are not more effective in treating CDI than standard antibiotic treatment with oral vancomycin or metronidazole
Treating CDI, placebo	NA	Low	Administration of a probiotic with live bacteria to treat CDI in critically ill patients increases risk for greater morbidity and mortality from fungemia without any known benefit
Treating recurrent CDI	Low Insufficient	Low NA	Fecal microbiota treatment is effective in treating recurrent CDI Data insufficient for patients with refractory CDI
Preventing CDI	NA	Low	Prebiotics and monoclonal antibodies are not more effective than placebo for primary prevention of CDI
Preventing recurrent CDI	Low Low NA	Low Low Moderate	Probiotics using lactobacillus strains are more effective than placebo for reducing recurrent CDI Probiotics using multiple organisms or *S. boulardii* are not more effective than placebo for reducing recurrent CDI Monoclonal antibodies are effective in preventing recurrence of CDI

CDI=*Clostridium difficile* infection

KQ1 – Diagnostic Tests

The literature has shown a strong shift from immunoassays to nucleic acid amplification tests, mirroring the evolution of clinical practice for diagnosis of CDI. Given the greatly increased published literature of diagnostic studies, we were disappointed at the lack of eligible studies of the impact of diagnostic tests on patient or health system outcomes; that is, does more accurate and expeditious diagnosis of CDI lead to improved outcomes for patients or measured improvement for health systems (with respect to cost of care, length of stay, or rates of CDI). Although some retrospective studies described changes in incidence and prevalence in an institution before and after implementation of a new testing strategy, these generally did not include verification of the reported incidence and prevalence with an acceptable reference standard and are thus difficult to interpret.

Interpreting the findings of the diagnostic testing evidence requires a nuanced approach. The pretest probability of CDI (and the severity of the CDI, if present) varies with patient

characteristics as well as clinical setting, with inpatient populations having higher prevalence and severity of disease. Determining the presence or absence of a given disease are not simply the obverse and reverse of each other, respectively. Testing strategies that emphasize sensitivity are better for ruling out disease while strategies that emphasize specificity are better at ruling in disease. In the inpatient setting—from which the vast majority of patients in the studies included in this report were drawn—a clinician's priority is on ruling out disease (and not treating for CDI) and thus a higher false positive rate is likely acceptable. From a Bayesian standpoint, a more sensitive test (with a lower negative likelihood ratio) is necessary to rule out disease to the same acceptable post-test probability than a less sensitive test applied to a lower pre-test probability. Since specificity was uniformly quite good across test classes, the difference in performance between classes of tests for CDI appears to be derived mostly from differences in sensitivity between classes. This has significant implications for testing strategy selection, since ensuring a negative test is accurately ruling out disease is likely of more clinical importance than ensuring that a positive test (in a patient with signs and symptoms consistent with CDI) represents true CDI. Thus, we believe the differences in sensitivity and negative likelihood ratios to have clinical importance.

There was substantial heterogeneity in the studies of diagnostic accuracy from both measured and unmeasured sources. The prevalence of CDI in the study population is one of the few consistently described population characteristics. The prevalence of CDI in the examined studies varied widely, between 6 and 48 percent. While sensitivity and specificity should theoretically not vary with prevalence, this is often the case in studies of diagnostic testing and future work should examine how the prevalence of CDI (likely a reflection of the population in a study and local testing behavior) influences the measured operating characteristics. There are many other undescribed clinical variables that may differ between populations and lead to heterogeneity. For example, training and implementation procedures generally were not described in detail. Lastly, the reference standard for each study was performed according to local protocols and, while used as the definition for the presence or absence of disease in the studies, may vary substantially, leading to different prevalence and operating characteristics. Unless a large, prospective trial is performed in which all reference standards (toxigenic culture and/or CCNA) are performed centrally to avoid local variation, these limitations are unlikely to be avoided in future studies.

The diagnostic studies included in this report included patients only with suspected CDI and thus the operating characteristics (that is, sensitivity and specificity) are defined in patients with suspected CDI, not general patients with diarrhea or healthy patients. Thus, these tests should only be interpreted in patients similar to patients enrolled in the included studies and clinicians must be aware of the prevalence of CDI in their own local population in determining whether they choose to employ a more sensitive or specific testing strategy.

For this update, we used a different approach to examine diagnostic tests, pooling studies by test class, since the selection of tests from within a test class for use at a certain institution will likely depend on both the operating characteristics of the test class and individual test as well other factors including cost and vendor preference. We found moderate to high evidence that NAAT tests (1 LAMP, 9 PCR) are highly sensitive, specific, and likely to function well as stand-alone diagnostic tests in hospital settings. All other diagnostic tests (8 Toxin A/B immunoassays, 4 GDH immunoassays, and 7 test algorithms) were high in sensitivity and/or specificity but the receiver operating characteristic curves did not compare as well to the NAAT receiver operating characteristic curve.

Test algorithms, intended to make the best of individual test strengths performed in series, did not perform as a class as well as NAAT tests. Clinical interest in test strategies has declined because of the issue of what to do when a positive initial test is followed by a negative test. Many clinicians will continue treating based on the first test because of the uncertainty (increased probability of CDI after the test), and because the test is usually ordered based on the clinician's pretest assessment of the patient's probability of CDI. Dichotomous, positive/negative results are easier for clinicians to interpret and require less laboratory followup.

NAAT tests come with a different set of concerns, including whether switching to NAAT will falsely inflate nosocomial CDI rates; these highly sensitive tests may identify people who are asymptomatic carriers or patients with diarrhea from a cause other than CDI. Since NAAT tests nearly approximate toxigenic culture in sensitivity and specificity, implementation of a NAAT-based testing strategy may lead to a higher observed prevalence/incidence compared to other testing strategies. Further research is required to determine if NAAT-based testing strategies lead to overtreatment for CDI in patients who are asymptomatic carriers or have diarrhea from another cause

To assume that one "best" test exists for all healthcare purposes is an oversimplification, especially with respect to populations with different pretest probabilities, as previously discussed. Further, clear delineation must be made between the most effective test characteristics for at-risk individuals who may benefit from CDI treatment versus for population surveillance or epidemiologic evaluation.

KQ2 – Prevention

The prevention literature remained generally low-strength, and little evidence connects prevention strategies directly to patient-related outcomes such as CDI incidence. Studies of transmission interruption techniques were often excluded due to lack of patient-related outcomes (they used swabbing and culturing to assess the presence of *C. difficile* organisms or spores). However, we did identify some small updates to the original review. Low-strength evidence supports a few new transmission interruption techniques: bathing patients with chlorhexidine gluconate and handwashing campaigns. Low-strength evidence also suggests that prevention programs are sustainable in the long-term. However, it remains difficult from a research perspective to definitively state that bundled, multicomponent interventions are effective, as each remains relatively unique to the specific location and the components included in that bundle. The information is still insufficient to answer which components are essential or what might be added.

Low-strength evidence continues to support antibiotic prescribing practices. Again, none of the studies explicitly addressed potential harms of changes in antibiotic use policy, such as the possibility that preferred drugs will be less effective than the drugs physicians are discouraged from using, or that preferred antimicrobials might have greater costs or greater toxicities unrelated to CDI.

KQ3 – Standard Treatment

Three new studies of standard treatment raised confidence in several findings from the original review. We increased strength of evidence from moderate to high for vancomycin as a more effective agent than metronidazole for CDI, with moderate-strength evidence of the effect regardless of severity. Current treatment guidelines from the Infectious Diseases Society of America (IDSA) support vancomycin as the drug of choice for severe CDI, and metronidazole as

the drug of choice for mild to moderate CDI.[64] This review's finding should prompt a reconsideration of the preferred agent for mild to moderate CDI. This is especially true in light of decreased concerns about the emergence of vancomycin-resistant enterococci during oral vancomycin therapy and a decrease in the price differential between metronidazole and vancomycin.

A second important finding is continuing moderate-strength evidence that fidaxomicin is similar to vancomycin for the initial cure of CDI, and increased strength of evidence for fidaxomicin as superior for the prevention of recurrent CDI. Since the desired outcome with CDI treatment is cure of the initial illness without subsequent recurrence, this finding ought to prompt consideration of fidaxomicin for the initial treatment of CDI. This is especially relevant to the treatment of CDI, since each episode of recurrence increases the likelihood of further episodes. Since fidaxomicin was licensed after publication of the most recent IDSA guidelines, they include no mention of fidaxomicin. Accordingly, its role in treating CDI has been a topic of considerable discussion. A recent cost-benefit analysis concluded that the per-course price of fidaxomicin would need to decrease by more than 10-fold in order to make such use cost-effective.[65] The current high cost of fidaxomicin prompted its manufacturer to seek and obtain a new technology add-on payment from the Centers for Medicare and Medicaid services. This add-on provides hospitals additional payment to offset the fidaxomicin's high cost. Future guidelines will hopefully give clinicians guidance as to how to best use this agent to maximize the value seen in terms of reduced episodes of recurrent CDI.

A final updated finding is that in the observational study of intravenous metronidazole verses oral metronidazole and vancomycin, intravenous metronidazole performed significantly worse than either oral drug. This finding should be interpreted with caution given the observational nature of the study and the significant possibility of confounding. Since this finding largely confirms current clinical practice, it will not likely have a major impact on the treatment of patients with CDI.

KQ4 – Adjunctive Treatments

Adjunctive treatments in the updated literature have largely focused on restoring the colonic microbiome for the prevention of subsequent CDI, although a few explored different mechanisms such as toxin-binding (tolevamer) and direct antimicrobial properties (lactoferrin). The diverse bacterial species residing in the human gut, commonly referred to as the colonic micobiome, provide host resistance to infection by *C. difficile*. Several factors, such as antimicrobial use and chemotherapy, disrupt the diversity of the colonic microbiome and lower the resistance to CDI. Antimicrobials are effective in treating CDI, but also disrupt the colonic biodiversity and do not address the necessary repopulation of these organisms. These changes make the host susceptible to recurrent episodes of CDI. Probiotics aim to recolonize the intestinal flora with nonpathogenic bacteria, while FMT involves the transfer of the entire microbiome from one individual or a pool of donors to the host.

Low-strength evidence supports FMT as a promising therapy for recurrent CDI. Since our original review, numerous studies have addressed FMT for the treatment of recurrent CDI, including one small unblinded RCT comparing FMT to two vancomycin-based control groups, one small RCT comparing two different modalities of administration for FMT, and several case series. The case series ranged from small to medium size and provide a cumulative experience with FMT of 648 individuals, with reported success rates from 70 – 100 percent. However, the high probability of publication bias and the lack of control groups are major limitation. The data

from the RCT comparing FMT to vancomycin is encouraging, demonstrating a significant benefit for FMT, although the study risk of bias is high. Specifically, participants and providers were unblinded, and the control groups had success rates from 23 to 31 percent, far lower than the 60 percent rates expected based on the sample size calculations published in the study protocol. Additionally, followup was limited in most studies; thus, the long-term consequences of FMT treatment are unknown.

Insufficient evidence exists for FMT for refractory CDI. In contrast to FMT for recurrent CDI—which is administered after a course of antimicrobial therapy has eliminated or greatly reduced symptoms of CDI, and whose main aim is to prevent subsequent recurrences—FMT for refractory CDI is administered to patients with ongoing symptoms of CDI despite antimicrobial therapy. Since the great majority of patients with CDI respond to initial antimicrobial treatment, studies of refractory CDI are inherently difficult.

The scientific and regulatory issues for FMT pose unique challenges, as there are no standard formulations, methods of quantifying, or assessing safety of stool. The composition of stool, and which constituents may be active in reducing recurrent CDI, is also currently unknown. Initial FDA guidance required an Investigational New Drug (IND) for any use of FMT. After significant public input, current FDA policy is to exercise enforcement discretion regarding IND requirements for FMT in specific situations. To proceed under enforcement discretion, FMT must be used to treat CDI not responding to standard therapies, and the treating physician must obtain an informed consent including, at a minimum, discussion of both the investigational nature of FMT and its potential risks (http://www.fda.gov/BiologicsBloodVaccines/GuidanceComplianceRegulatoryInformation/Guidances/Vaccines/ucm361379.htm). This guidance is subject to change as more evidence and experience accumulates on FMT, including the optimal route of FMT delivery. The sources of material for FMT are also variable, and include use of unrelated or related donors. No standard criteria exist for screening donors, but several guideline documents have been developed. The optimal team to administer and oversee an FMT program is uncertain, but may include gastroenterology, infectious diseases, pharmacy, infection control, nursing, and facility management.

The low-strength evidence supporting probiotics for the prevention of CDI is mixed, and somewhat contradictory. Preparations containing either *S. boulardii* alone or multiple organisms did not seem to significantly affect subsequent rates of CDI, whereas preparations containing only *lactobacillus* strains did significantly reduce rates of CDI. Notably, these studies aimed to examine probiotics for primary prevention of CDI among patients without a prior episode of CDI. Whether the findings apply to patients with a history of CDI (that is, to prevent recurrence of CDI) is unknown. Administering a course of probiotics to every patient taking antimicrobials would also be a rather substantial change in medical practice. The cost/benefit ratio of such a policy is unclear, based on the mixed and low-strength findings of this report, as is whether benefits could be conferred by ingesting probiotics in the form of yogurt, kefir, and other similar foods. Given the multitude of such foods available to consumers, the prospect of obtaining rigorous data on each seems unlikely.

Finally, rifaximin and lactoferrin were studied in separate small placebo controlled trials regarding their ability to prevent subsequent episodes of CDI. Rifaximin was given after a course of standard antimicrobial therapy for CDI in hopes of preventing recurrent CDI. In contrast, lactoferrin was given concomitantly to antibiotics prescribed for non-CDI indications in the

hopes of preventing an initial episode of CDI. In both studies, the investigational agent reduced the incidence of subsequent diarrhea, but not confirmed CDI.

The bulk of the new studies of adjunctive treatment for the prevention of subsequent CDI involve efforts to reconstitute the colonic microbiome with either FMT or probiotics. The supporting evidence is low-strength. The FMT studies show a large treatment effect, but are limited by methodological weaknesses. In contrast, the studies supporting probiotics demonstrate a less-impressive treatment effect. The FMT studies included patients with at least a single prior episode of CDI, and in many cases, multiple prior episodes. The probiotic trials, on the other hand, are examining primary prevention of CDI. Both primary and secondary prevention of CDI are important, particularly since the burden of CDI has significantly increased over the past 15 years.

Research Gaps

For diagnostic studies, in spite of the increased applicable evidence in this update, many differences persisted in each laboratories' CDI reference standard sensitivity and specificity, especially the relatively strict reference standard for CDI we specified for included studies. Reference standards are used to determine CDI prevalence, but no reference standard has perfect sensitivity and specificity. Even small differences in prevalence in a population may lead to markedly different predictive values.

The marked heterogeneity in the operating characteristics of the tests analyzed was puzzling. No tests have perfect operating characteristics; thus, we could not determine the clinical significance of the differences in operating characteristics between individual tests and classes of tests. Future studies should determine whether the differences in operating characteristics for the same proprietary test between laboratories are the result of patient/sample characteristics, prevalence of disease, test performance, reference standard performance, or other factors. The findings of these studies would likely be of significant pragmatic importance as new testing strategies are applied in health systems across the country. Best practices for testing that are independent of the manufacturer should be developed.

The criteria for future studies outlined in the previous report with a few modifications (in italics) should be applied to future multicenter studies: 1) Use the most clinically relevant reference test (which may be NAATs); 2) Use explicit clinical criteria to select patients and stool specimens to be tested; 3) Randomly assign patients to different diagnostic tests *(or perform and interpret multiple tests independently)*; and 4) Use key clinical outcomes as study endpoints are needed. Also, studies should prospectively determine how inconclusive results will be handled, and all samples should be included in the determination of operating characteristics and the result of each test (if tests are applied serially) made available for analysis.

For prevention, the main obstacle to research continues to be the contextual setting. To design and conduct studies with adequate comparators to allow for causal inference is certainly challenging. Nonetheless, the field would benefit from such work. Indeed, study designs did improve, attempting to use prospective data collection and interrupted time series. Further use of implementation science techniques may move the field forward. Additional studies of transmission interruption follow results past culturing room swabs to clinical outcomes such as CDI incidence would also be of benefit.

Future research needs for the treatment of the first episode of CDI include studies to identify subjects who derive the most benefit from fidaxomicin, and studies of new agents to further decrease the recurrence rate from the 14 percent observed with use of fidaxomicin. A few new

agents are currently under investigation. (See Appendix Table I1.) Recurrent CDI is difficult for both patients and clinicians to manage; thus, lowering the recurrence rate as much as possible is a high clinical priority. Finally, since both the largest RCT of metronidazole verses vancomycin and pooled data from all such trials indicate that vancomycin is superior to metronidazole for the initial cure of CDI, further studies comparing these agents are not likely to be clinically useful.

Adjunctive treatments for CDI need more research. FMT is particularly challenging to research. It involves highly complex microbial mixtures that vary from donor to donor. Additionally, several delivery routes are used, including instillation of donor feces into the upper gastrointestinal (GI) tract via nasogastric or naso-jejunal tubes or in an oral capsule, instillation of feces into the distal colon via enema, or instillation in the entire colon via colonoscopy. Numerous Phase 2 studies on safety and efficacy for FMT can be found on ClinicalTrials.gov. Only one study has compared the safety and efficacy of various routes; the authors compared FMT delivery via colonoscopy and nasogastric tube and reported no difference in efficacy of preventing recurrent CDI.[55] Future research should focus on adequately powered, controlled, and blinded RCTs assessing FMT, and include long-term followup; several such studies are already registered. (Appendix Table I1)

Further research is also needed to sort out the unexpectedly contradictory findings for probiotics. This is a challenging topic, since the human gut ecology is a complex system. Since food and beverages can also be significant sources of probiotics, establishing clear comparator groups can be difficult. Further information generated by the human biome research initiative may help inform this area.

A randomized trial of different therapies for refractory patients, including FMT, would also advance the field.

Limitations

This review has several limitations. In keeping with the original review, most diagnostic studies included in this update enrolled samples from patients at risk for or with symptoms consistent with CDI. However, some studies included unformed specimens only regardless of whether testing for CDI was requested by the patients' clinician. Studies generally did not describe the clinical characteristics of the patients from whom fecal samples were obtained for inclusion, making it difficult to determine the applicability of findings. Further, we could not determine the impact of enrolling nonconsecutive samples on the measured operating characteristics of a certain diagnostic test. We cannot exclude the possibility that a study with nonconsecutive sample of patients could systematically entrain bias if there were characteristics that led to samples being included and others excluded, such as volume of stool, variability of testing practices in certain wards, or other characteristics.

Requiring patient-centered outcomes such as CDI incidence for prevention studies resulted in the exclusion of several transmission interruption studies. Some decisionmakers may be willing to use studies that examined intermediate outcomes such as the number of cultures obtained from swabs. However, we encountered no literature directly tying numbers of cultures to actual CDI incidence and thus could not infer clinical meaning from a reduction in cultured swabs.

Pooling diagnostic tests by test class resulted in heterogeneity for most test classes. We examined the heterogeneity for pooled individual tests with sufficient numbers of studies and found significant heterogeneity in these meta-analyses as well. Thus, we deemed the gain in information by pooling test classes worth the cost of the added uncertainty from the heterogeneity.

Conversely, pooled meta-analyses for probiotics studies showed low heterogeneity, even though we pooled liberally based on the probiotic strain(s) included in each study. Pooling used a conceptual basis, representing only one possible way of categorizing the probiotic interventions.

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Abbreviations

AHRQ	Agency for Healthcare Research and Quality
CCNA	Cell cytotoxicity neutralization assay
CDI	*Clostridium difficile* infection
CENTRAL	Cochrane Central Register of Controlled Trials
CER	Comparative Effectiveness Review
CI	Confidence interval
FMT	Fecal microbiota transplantation
GDH	Glutamate dehydrogenase
GI	Gastrointestinal
ICTRP	International Controlled Trials Registry Platform
IDSA	Infectious Disease Society of America
IND	Investigational New Drug
LAMP	Loop mediated isothermal amplification
MRSA	Methicillin-resistant staphylococcus aureus
NAAT	Nucleic acid amplification tests
PCR	Polymerase chain reaction
PICOTS	Population, Interventions, Comparators, Outcomes, Timing, Settings
RCT	Randomized controlled trial
RD	Risk difference
ROC	Receiver operating characteristic
RR	Risk ratio
SIP	Scientific information packet
SMD	Standard mean difference
WMD	Weighted mean difference