CME

Clostridium difficile–associated colitis

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ABSTRACT

OBJECTIVE To review the basic microbiology, pathogenesis of disease, and diagnosis of the nosocomial pathogen *Clostridium difficile* and to examine therapies recommended by the Canadian Task Force on Preventive Health Care.

QUALITY OF EVIDENCE MEDLINE was searched using MeSH headings. Controlled trials for therapy were sought, but case-control studies and observational reviews were included.

MAIN MESSAGE *Clostridium difficile* causes approximately 20% of cases of diarrhea associated with antibiotics, including clindamycin and the second- and third-generation cephalosporins. Diarrhea is usually mild, but can be severe; extreme cases develop toxic megacolon. Diagnosis is dependent on demonstrating presence of clostridial toxin in stool specimens or of pseudomembranes through sigmoidoscopy. First-line therapy for *C difficile* diarrhea is restricted to metronidazole. Second-line therapy for treatment failure is vancomycin. For relapse, a second course of metronidazole is recommended; tapering courses of vancomycin and probiotics are used for multiple recurrences.

CONCLUSION *Clostridium difficile* is an important nosocomial pathogen requiring prudent use of antibiotics and strict infection-control policies to prevent large health care costs.

RÉSUMÉ

OBJECTIF Faire le point sur la microbiologie, la pathogénicité et le diagnostic de l'agent pathogène nosocomial *Clostridium difficile* et examiner les traitementss recommandés par le Groupe de travail canadien sur la médecine préventive.

QUALITÉ DES PREUVES MEDLINE a été répertorié à l'aide de mots clés MeSH. On a retenu des essais thérapeutiques contrôlés, mais aussi des études cas-témoins et des études d'observation.

PRINCIPAL MESSAGE Le *C.difficile* est responsable de 20% environ des cas de diarrhée associés à des antibiotiques, notamment la clindamycine et les céphalosporines de troisième génération. La diarrhée est généralement bénigne, mais elle peut être sévère, avec complication de mégacôlon toxique dans les cas extrêmes. Le diagnostic repose sur la découverte de la toxine du *Clostridium* dans les selles ou de pseudomembranes à la sigmoïdoscopie. Le métronidazole est utilisé seul comme traitement initial, avec la clindamycine comme second recours. En cas de rechute, on recommande une nouvelle cure au métronidazole. Les rechutes multiples exigent l'utilisation de vancomycine et d'agents probiotiques.

CONCLUSION Le *C. difficile* est un important agent pathogène nosocomial qui requiert une antibiothérapie prudente et de strictes mesures de contrôle d'infection afin éviter des coûts de santé élevés.

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Antibiotic use is now commonplace both within hospitals and in the community at large. Diarrhea is a common complication of antibiotic therapy: quinolones and macrolides are associated with rates of 2% to 5%, while other agents (such as amoxicillin-clavulanate and cefixime) lead to diarrhea in 10% to 25% of treated patients (**Table 1**).[1]

Table 1. Common antibiotics associated with *C difficile* diarrhea

MOST COMMON
Clindamycin
Penicillin derivatives (especially amoxicillin-clavulanate)
Cephalosporins (especially third generation)
LESS COMMON
Macrolides
Quinolones
Tetracyclines
RARE
Metronidazole
Vancomycin

Approximately 15% to 20% of cases of antibiotic-associated diarrhea are caused by a single bacterial entity, *Clostridium difficile*.[2,3] *Clostridium difficile* infections are an increasing problem; *C difficile* is recognized as the most common nosocomial gastrointestinal infection. Originally isolated in 1935, *C difficile* was initially thought to be a component of normal flora, and was not identified as a pathogen until the 1970s when colitis associated with clindamycin use was further investigated. In this update we review *C difficile* disease emphasizing pathophysiology, diagnosis, and treatment from family physicians' perspective.

Quality of evidence

A MEDLINE search was conducted using the key words *Clostridium difficile*, pseudomembranous colitis, and antibiotic-associated diarrhea. The search was limited to English-language articles, clinical trials, and review articles. Publications thus selected were then reviewed for additional pertinent references, and a final list of original articles was compiled for inclusion.

Microbiology and pathophysiology

Clostridium difficile is an anaerobic Gram-positive bacillus capable of spore production. These spores are heat resistant and can survive in the environment for many months despite desiccation and exposure to disinfectants. There have been reports of viable spores being found on clothing and hospital furniture and equipment. Thus, these spores contribute to exogenous exposure to *C difficile* via the fecal-oral route. Spores survive ingestion to germinate in the colon to form vegetative bacilli capable of growth and toxin production.

Pathogenic strains of *C difficile* cause diarrhea and colitis via toxin production.[4] Two major toxins have been identified: toxin A is a 308kD enterotoxin, and toxin B is a 269kDa cytotoxin. Both toxins are capable of stimulating production of proinflammatory cytokines[4] that have been implicated in the pathogenesis of pseudomembranous colitis.[3] The toxins act by altering the regulation of cytoskeletal protein, resulting in cell rounding and ultimately cell death.[5] Animal models demonstrate that toxin A induces epithelial desquamation and increased mucosal permeability leading to increased fluid secretion.[6] Toxin B lacks significant enterotoxic effects in animal models and was not considered a substantial contributor to human disease. Recent *in vitro* data using human colonic epithelial cell lines suggests, however, that toxin B is 10 times more potent at inducing colonic injury than toxin A.[7] This finding is consistent with clinical evidence indicating that toxin A–negative—toxin B–positive strains can account for nosocomial disease, as evidenced by a recent outbreak in Winnipeg, Man.[8]

In addition to toxin production, *C difficile* can harbour specific antibiotic-resistant gene cassettes such as the *ermB* gene, which encodes a 23S ribosomal methylase responsible for resistance to

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macrolide-lincosamide-streptogrammin (MLS) antimicrobial agents. This MLS marker has been identified in several outbreaks of *C difficile* in which exposure to the antibiotic clindamycin, a derivative of the lincosamide group, was shown to be a trigger.[9]

Clostridium difficile requires a perturbation in the normal colonic flora microenvironment for overgrowth, increased toxin production, and thus clinical disease to occur.[2] Indigenous intestinal flora exert a protective effect referred to as colonization resistance, which generally limits colonization by pathogenic microorganisms. Antibiotic therapy disrupts these protective flora allowing colonization and overgrowth by *C difficile.*

Clinical manifestations

Clostridium difficile disease presents in a variety of ways, ranging from asymptomatic carrier status to moderate diarrhea and life-threatening pseudomembranous colitis.[10] Symptoms usually manifest as profuse diarrhea that is watery or mucous, sometimes accompanied by abdominal pain and fever (**Table 2**). Leukocytosis is a common indicator of *C difficile* infection; white blood cell counts greater than 30.0 x 10^9/L can occur[11] (**Table 2**). Extraintestinal manifestations are rare, but can include cellulitis, bacteremia, abscess formation in the viscera, and reactive arthritis.[12]

Patients with more severe disease might have marked abdominal pain and distention, sometimes accompanied by peritoneal signs. In addition to the diarrhea, systemic features of anorexia, fever, dehydration, hypoalbuminemia, and electrolyte disturbances can appear[3,13] (**Table 2**). Severe pseudomembranous colitis can have serious consequences[14]; patients can present requiring emergency care, acutely ill with life-threatening colitis.[15] Radiographic investigations can show evidence of toxic megacolon with colonic dilation on plain film (**Figure 1A**). Computed tomography findings include colonic wall thickening, pancolitis, and pericolonic inflammation[16] (**Figure 1B**).

Endoscopic evaluation can show diagnostic pseudomembranes or lesions suggestive of nonspecific colitis, such as erythema, edema, and friability.[17] Characteristic lesions are yellow plaques 2 to 10 mm in diameter with normal mucosa interposed (**Figure 1C**). In patients with more severe disease, these lesions enlarge to cover substantial portions of inflamed mucosa, but can be easily stripped off (thus the term pseudomembrane).[13]

Table 2. Clinical presentations

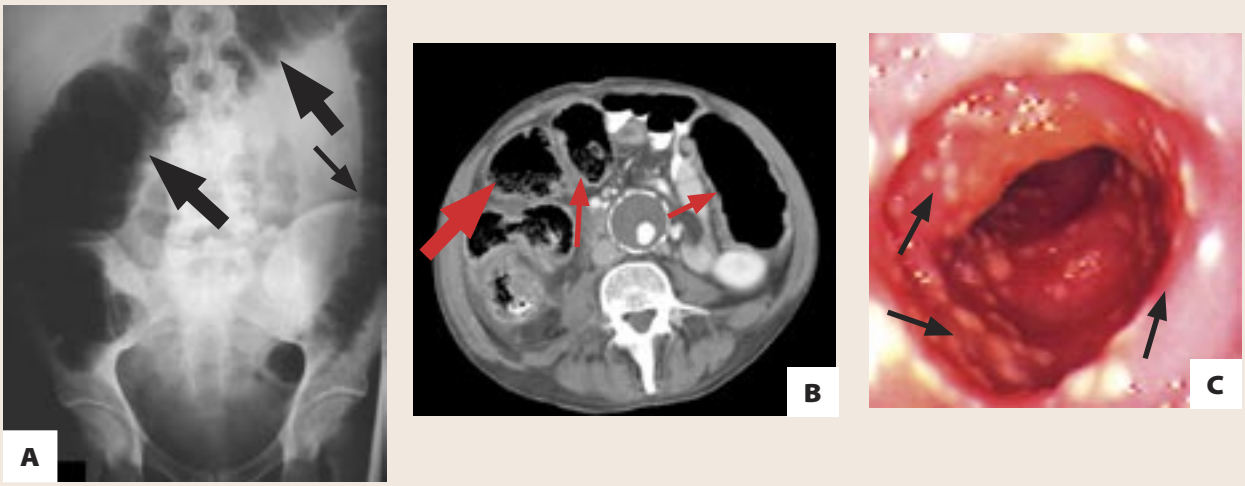
MILD
Diarrhea
Abdominal cramping
Tenesmus
Low-grade leukocytosis
MODERATE
Leukemoid reaction
Fever
Dehydration
Nausea, vomiting
Abdominal tenderness
SEVERE
Sepsis or shock
• Acidosis
• Multisystem organ failure
Tachycardia
Acute abdomen (colonic perforation)
Toxic megacolon
Ascites
Paralytic ileus
Hypoalbuminemia
Diarrhea can actually lessen in severe disease

Surgery is usually unnecessary, except for 0.4% to 5% of patients who fail to respond to medical therapy or have impending multiorgan failure, toxic dilation, or perforation.[18,19] The procedure of choice is a subtotal colectomy and ileostomy. Other approaches, including segmental resections and diverting stomas, have higher associated failure rates and complications.[20] Mortality for mild cases in collected series ranges from 1.1% to 3.8%; mortality for severe cases requiring surgery can exceed 30%.[18-20]

Epidemiology

While neonatal and infant carriage rates of *C difficile* are as high as 50%,[21] colonization is usually asymptomatic.

Figure 1. Features of *C difficile* colitis: *A) Plain film of abdomen showing bowel wall thickening, loss of haustral markings (thin arrow) and dilation of the ascending and transverse colon (thick arrow); B) Computed tomographic scan of abdomen showing colonic dilation (thin arrows) and bowel wall thickening with stranding (thick arrow); C) Endoscopic view of classic* C difficile*–associated pseudomembranous colitis (arrows mark pseudomembranes).*



Colonization among healthy adults is low (0 to 3%), reflecting exogenous acquisition as a precursor for disease.[14] Studies have implicated hospital environments for nosocomial acquisition of *C difficile*. In one study, 21% of patients acquired *C difficile* after admission with a median of 12 days between admission and testing positive for *C difficile;* most (63%) remained asymptomatic.[22] Other studies have shown that length of stay influences acquisition; the incidence of *C difficile* increases greatly after 4 weeks in hospital.[23] McFarland et al[22] identified positive environmental cultures in 49% of rooms occupied by symptomatic patients, in 29% of rooms occupied by asymptomatic patients, and in only 8% of rooms occupied by patients whose *C difficile* cultures were negative. These data clearly show the importance of person-to-person transmission of *C difficile* within hospitals. Acquisition is more likely with prolonged exposure to contaminated environments. The community rate of *C difficile* is estimated to be between 7 and 12 cases per 100000 person-years.[24] Incidence for nonepidemic hospital settings ranges from 0.1 to 30 per 1000 patients.[14]

Risk factors

Antibiotic exposure is the single most important risk factor, contributing to altered gut flora and allowing overgrowth of *C difficile*. Disease can occur after a few days of antibiotic therapy or up to 2 months after use.[13] Clindamycin has been identified as a preeminent agent in this regard, a finding confirmed by recent epidemics where clonal strains of *C difficile* resistant to clindamycin by virtue of the *ermB* gene have been isolated.[9] Other antibiotics commonly found to predispose patients to infection include the second- and third-generation cephalosporins and penicillins.[25] Less common agents include the quinolones, sulfonamides, and metronidazole.[3] In addition to antibiotics, cancer chemotherapy is a risk factor.[26] Other important risk factors are increased age and severity of underlying illness.

Diagnosis

Laboratory testing serves to confirm *C difficile*–associated disease in patients with underlying risk factors and clinical profile. A variety of tests have been developed for testing diarrheal stools for *C difficile*, and their availability depends on local practice in various health regions.

Enzyme-linked immunoassay for toxin. These rapid tests use monoclonal antibodies to detect toxin, and are probably the most widely used assays.

Commercial kits are available that detect toxin A, toxin B, or both toxins.[27,28] These tests have lower sensitivity (63%-99%) than cytotoxin culture, but high specificity (85%-100%).[29] If test results are reported negative, sending one or two additional stools on subsequent days could improve sensitivity.[13] In addition, if toxin A alone is tested, diarrhea secondary to toxin A–negative—toxin B–positive strains will be missed.[8,30]

Cytotoxicity assay. This test is positive when stool filtrate inoculated onto cell culture monolayer demonstrates cytopathic effect due to the presence of *C difficile* toxin. It remains the criterion standard of detection. Sensitivity and specificity are 67% to 100% and 85% to 100%, respectively.[29] The test, however, is relatively expensive; it requires 24 to 48 hours for completion; and inactivation of toxins during transport, through dilution of sample, and through age of the cell line gives rise to false-negative results.

Other methods. Latex agglutination–based assays recognize the enzyme glutamate dehydrogenase but lack sensitivity. Stool culture for *C difficile* is sensitive, but will also detect nontoxigenic strains.[3] Genetic analysis via polymerase chain reaction is available, but confined to research settings. Endoscopy should be reserved for when a patient's condition necessitates rapid diagnosis or when other diagnoses are being considered. Colonoscopy can detect cases not apparent during sigmoidoscopy because *C difficile* infection can involve the transverse colon or ascending colon cecum without evidence of involvement of the more distal segments of the colon.

False-negative results can occur in any of these assays, so if the clinical suspicion is high and the assay is negative, either the test should be repeated or sigmoidoscopy or colonoscopy should be considered. This is critical, for we have seen several cases where the initial assay is negative and empiric *C difficile* therapy is stopped or the diagnostic possibility of *C difficile* is abandoned, resulting in delayed treatment and unfortunately serious adverse outcomes for some patients.

Treatment

Once *C difficile* diarrhea is diagnosed, inciting antibiotic therapy must be discontinued (if possible). Antidiarrheal agents should be avoided and narcotics minimized in order to prevent colonic stasis and decreased toxin clearance. Volume resuscitation is necessary for dehydrated patients. Specific treatment involves antibiotic regimens directed at *C difficile*.

Metronidazole. Metronidazole is recommended as first-line therapy for *C difficile* diarrhea in nonpregnant adults. It is inexpensive and generally well tolerated; side effects include a metallic taste, nausea, peripheral neuropathy, and disulfiram effect. Clinical experience with the use of metronidazole is extensive: a review of 908 cases over 10 years at an institution showed that 70% of patients were treated in this fashion, with a 1% intolerance rate and 2% failure rate.[31] Some prospective randomized trials have involved metronidazole. In one study 94 patients with *C difficile* diarrhea were randomized to 250 mg of metronidazole four times daily or 500 mg of vancomycin for 10 days. Results indicated similar responses in both arms; two patients taking metronidazole had treatment failures.[32] In another similar trial the cure rate between the vancomycin and metronidazole arms was identical (94%),[33] giving level I evidence for metronidazole use.

Vancomycin. Oral vancomycin is not well absorbed from the gastrointestinal tract and has intracolic effect, with response rates equal to that of metronidazole and superior to other therapies.[32-35] There is level I evidence for vancomycin use. Initial studies used dosages of 500 mg four times daily for 10 days; however, a dose of 125 mg four times daily has similar efficacy.[36] Vancomycin is second-line therapy and, because of its cost and the need to prevent development of vancomycin resistance in species such as *Enterococcus*, is reserved for patients whose treatment with metronidazole fails to provide benefit.[37]

Other therapies. Other antibiotics shown to be effective include bacitracin[34,38] at dosages of 80 000 to 100 000 U daily, although it is less effective than

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vancomycin. Teicoplanin, another oral glycopeptide, has similar efficacy to vancomycin[33,35] (level II evidence), but is not easily available. Fusidic acid has been used in a few patients[33,39] but had less success in terms of overall cure, and relapse rates were higher.

In patients who develop ileus, combination therapy with oral and intravenous agents has been suggested (level III evidence).[40] Some patients have been treated with oral antibiotics administered through nasogastric tube and metronidazole administered intravenously. Some evidence suggests that bactericidal concentrations can be reached in the colon with intravenous metronidazole, but the treatment route of choice is still by mouth.[41] There are also reports of using vancomycin-retention enemas and of infusing vancomycin into the colon via a rectally placed catheter for patients unable to tolerate oral medications.

Asymptomatic patients. Epidemiologic data demonstrate that *C difficile* can be acquired asymptomatically within hospitals and subsequently transmitted to other patients.[22,23] Evidence that these patients should be isolated is lacking, however, and as treatment with metronidazole and vancomycin does not reliably eradicate spore carriage in asymptomatic carriers, guidelines from both the American College of Gastroenterology and the Society for Hospital Epidemiology of America recommend against testing stool samples from asymptomatic patients, including posttreatment tests for cure (level III evidence), and recommend against treating asymptomatic patients (level I).[13,29]

Recurrent* C difficile *diarrhea. Approximately 15% to 35% of patients (mean 20%) will have recurrent disease, despite initial treatment.[41] This could be from reinfection or from germination of residual spores within the colon. There is no evidence that recurrent infections cause more severe disease.[41] The first relapse should be treated with a repeat course of metronidazole[13] (level III evidence).

Observational data indicate only 8% of cases had more than one relapse.[31] For cases of multiple relapse, treatment strategies vary, but supportive evidence is poor. Tedesco et al[42] published a series of 22 patients treated with a tapering oral vancomycin schedule in which the 125-mg dose was administered four times daily but reduced to 125 mg twice daily and then again by half on a weekly basis until pulse doses of 125 mg of vancomycin were administered every 3 days for 2 weeks[42] (level III evidence). The tapering treatments were aimed at eliminating germinating spores over an extended period. This regimen has received some support in a post-hoc analysis of the placebo arm of a randomized controlled trial for *Saccharomyces boulardii* used to treat recurrent cases: placebo cases treated with tapering courses of vancomycin had significantly fewer recurrences.[43] Vancomycin and rifampin combination therapy was shown to be beneficial in a small uncontrolled study of seven patients[44] (level II evidence).

Biologic agents. In a placebo-controlled trial, 124 patients were randomized to receive either standard antibiotic therapy, or antibiotics and *Saccharomyces boulardii*, a yeast previously shown to be beneficial in animal models. Treatment with the yeast for 4 weeks was shown to reduce recurrence rates from 65% to 35% in patients with recurrent disease[45] (level I evidence). Other agents that have been studied include *Lactobacillus GG*, although at present evidence is restricted to a case series of five patients and to preliminary data from a controlled study[46] (level II evidence). In cases of continued relapse, other therapies, such as rectal instillation of a mixture of anaerobic bacteria or rectal enemas of feces from healthy relatives, have been used to restore colonic flora.[47]

When to get help

There are no clear-cut guidelines on when to refer patients to specialists or when patients should be hospitalized. Because many episodes of *C difficile* colitis likely resolve without therapy or even a visit to a physician, we believe that many, if not most, cases can adequately be handled by primary care physicians in the community. We suggest referring patients or seeking hospital admission if they:

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- have trouble maintaining their volume status;
- have any signs of systemic toxicity, such as high fevers, marked elevations in white cell counts, or bandemia;
- have severe abdominal pain or tenderness;
- have any peritoneal signs;
- have any features of toxic megacolon; or
- have any signs of sepsis.

Referral should also be considered when diagnosis is in question, for those who fail to respond to therapy, and for those with other serious comorbid illnesses. If recurrent disease has relapsed after two or more courses of therapy, referral to a specialist should be considered.

Conclusion

Clostridium difficile has become well recognized as a major nosocomial pathogen associated with antibiotic use within hospitals and communities. Disease due to *C difficile* is responsible for substantial strain on the hospital system by increasing patients' length of stay and increasing costs. Diagnosis should be considered if patients have diarrhea and a history of antibiotic exposure. Toxin detection assays remain the first step in proving infection; however, sigmoidoscopy should be considered if patients have possible alternative diagnoses or if assay results are negative but clinical suspicion is high. Prevention of infection, by prudent restriction of antibiotic use, strict adherence to hand washing, disinfection, and infection control precautions for infected patients, is crucial. Antibiotic therapy for first-episode infections is well established; however, treatment of relapsed cases is less well defined and multiple relapses could require lengthy therapy.

Acknowledgment

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Competing interests

None declared

EDITOR'S KEY POINTS

- Diarrhea is a common complication of antibiotic treatment: associated with about 2% to 5% of cases treated with quinolones and macrolides and with 10% to 25% of cases treated with clindamycin, clavulin, and third-generation cephalosporins.
- Clinical manifestations of *C difficile* range from asymptomatic carrier state to the usual diarrhea and abdominal cramps, to sepsis, shock, and toxic megacolon.
- Rates of *C difficile* are much higher among in-hospital patients; risk increases with length of stay. Diagnosis is based on testing for A and B toxin.
- Treatment includes discontinuing the offending antibiotic, avoiding antidiarrheal agents, and supportive care for toxic cases. Metronidazole is first-line treatment with vancomycin as backup. Recurrence is common and patients should be re-treated with metronidazole first.

POINTS DE REPÈRE DU RÉDACTEUR

- La diarrhée est une complication fréquente de l'antibiothérapie : elle survient chez environ 2-4% des patients traités par les quinolones et les macrolides et dans 10-25% des cas traités par la clindamycine, le clavulin et les céphalosporines de troisième génération.
- Les manifestations cliniques du *C. difficile* vont de l'état de porteur sain au mégacôlon toxique, en passant par la diarrhée habituelle avec crampes abdominales, la septicémie et le choc.
- Le *C. difficile* est beaucoup plus fréquent chez les patients hospitalisés, le risque augmentant avec la durée du séjour. Le diagnostic repose sur la présence des toxines A et B.
- Le traitement inclut l'arrêt de l'antibiotique responsable, l'abstention d'agents antidiarrrhéiques et le support de l'état général en cas de toxicité. Le métronidazole est employé en premier recours et la vancomycine en second. Les rechutes sont fréquentes et exigent une nouvelle cure de métronidazole.

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References

1. Bartlett JG. Clinical practice. Antibiotic-associated diarrhea. *N Engl J Med* 2002;346(5):334-9.
2. Bartlett JG. *Clostridium difficile*: history of its role as an enteric pathogen and the current state of knowledge about the organism. *Clin Infect Dis* 1994;18(Suppl 4):S265-72.
3. Kelly CP, Pothoulakis C, LaMont JT. *Clostridium difficile* colitis. *N Engl J Med* 1994;330(4):257-62.
4. Warny M, Keates AC, Keates S, Castagliuolo I, Zacks JK, Aboudola S, et al. p38 MAP kinase activation by *Clostridium difficile* toxin A mediates monocyte necrosis, IL-8 production, and enteritis. *J Clin Invest* 2000;105(8):1147-56.
5. Pothoulakis C. Pathogenesis of *Clostridium difficile*-associated diarrhoea. *Eur J Gastroenterol Hepatol* 1996;8(11):1041-7.
6. Mitchell TJ, Ketley JM, Burdon DW, Candy DC, Stephen J. Biological mode of action of *Clostridium difficile* toxin A: a novel enterotoxin. *J Med Microbiol* 1987;23(3):211-9.
7. Riegler M, Sedivy R, Pothoulakis C, Hamilton G, Zacherl J, Bischof G, et al. *Clostridium difficile* toxin B is more potent than toxin A in damaging human colonic epithelium in vitro. *J Clin Invest* 1995;95(5):2004-11.

8. Alfa MJ, Kabani A, Lyerly D, Moncrief S, Neville LM, Al-Barrak A, et al. Characterization of a toxin A–negative, toxin B–positive strain of *Clostridium difficile* responsible for a nosocomial outbreak of *Clostridium difficile*–associated diarrhea. *J Clin Microbiol* 2000;38(7):2706-14.
9. Johnson S, Samore MH, Farrow KA, Killgore GE, Tenover FC, Lyras D, et al. Epidemics of diarrhea caused by a clindamycin-resistant strain of *Clostridium difficile* in four hospitals. *N Engl J Med* 1999;341(22):1645-51.
10. Gerding DN. Disease associated with *Clostridium difficile* infection. *Ann Intern Med* 1989;110(4):255-7.
11. Wanahita A, Goldsmith EA, Musher DM. Conditions associated with leukocytosis in a tertiary care hospital, with particular attention to the role of infection caused by *Clostridium difficile*. *Clin Infect Dis* 2002;34(12):1585-92.
12. Jacobs A, Barnard K, Fishel R, Gradon JD. Extracolonic manifestations of *Clostridium difficile* infections. Presentation of 2 cases and review of the literature. *Medicine* (Baltimore) 2001;80(2):88-101.
13. Fekety R. Guidelines for the diagnosis and management of *Clostridium difficile*–associated diarrhea and colitis. American College of Gastroenterology, Practice Parameters Committee. *Am J Gastroenterol* 1997;92(5):739-50.
14. Kyne L, Farrell RJ, Kelly CP. *Clostridium difficile*. *Gastroenterol Clin North Am* 2001;30(3):753-77, ix-x.
15. Triadafilopoulos G, Hallstone AE. Acute abdomen as the first presentation of pseudomembranous colitis. *Gastroenterology* 1991;101(3):685-91.
16. Cleary RK. *Clostridium difficile*–associated diarrhea and colitis: clinical manifestations, diagnosis, and treatment. *Dis Colon Rectum* 1998;41(11):1435-49.
17. Gebhard RL, Gerding DN, Olson MM, Peterson LR, McClain CJ, Ansel HJ, et al. Clinical and endoscopic findings in patients early in the course of *Clostridium difficile*–associated pseudomembranous colitis. *Am J Med* 1985;78(1):45-8.
18. Synnott K, Mealy K, Merry C, Kyne L, Keane C, Quill R. Timing of surgery for fulminating pseudomembranous colitis. *Br J Surg* 1998;85(2):229-31.
19. Jobe BA, Grasley A, Deveney KE, Deveney CW, Sheppard BC. *Clostridium difficile* colitis: an increasing hospital-acquired illness. *Am J Surg* 1995;169(5):480-3.
20. Morris JB, Zollinger RM Jr, Stellato TA. Role of surgery in antibiotic-induced pseudomembranous enterocolitis. *Am J Surg* 1990;160(5):535-9.
21. Larson HE, Barclay FE, Honour P, Hill ID. Epidemiology of *Clostridium difficile* in infants. *J Infect Dis* 1982;146(6):727-33.
22. McFarland LV, Mulligan ME, Kwok RY, Stamm WE. Nosocomial acquisition of *Clostridium difficile* infection. *N Engl J Med* 1989;320(4):204-10.
23. Clabots CR, Johnson S, Olson MM, Peterson LR, Gerding DN. Acquisition of *Clostridium difficile* by hospitalized patients: evidence for colonized new admissions as a source of infection. *J Infect Dis* 1992;166(3):561-7.
24. Levy DG, Stergachis A, McFarland LV, Van Vorst K, Graham DJ, Johnson ES, et al. Antibiotics and *Clostridium difficile* diarrhea in the ambulatory care setting. *Clin Ther* 2000;22(1):91-102.
25. Nelson DE, Auerbach SB, Baltch AL, Desjardin E, Beck-Sague C, Rheal C, et al. Epidemic *Clostridium difficile*–associated diarrhea: role of second- and third-generation cephalosporins. *Infect Control Hosp Epidemiol* 1994;15(2):88-94.
26. Anand A, Glatt AE. *Clostridium difficile* infection associated with antineoplastic chemotherapy: a review. *Clin Infect Dis* 1993;17(1):109-13.
27. Lyerly DM, Neville LM, Evans DT, Fill J, Allen S, Greene W, et al. Multicenter evaluation of the *Clostridium difficile* TOX A/B TEST. *J Clin Microbiol* 1998;36(1):184-90.
28. Merz CS, Kramer C, Forman M, Gluck L, Mills K, Senft K, et al. Comparison of four commercially available rapid enzyme immunoassays with cytotoxin assay for detection of *Clostridium difficile* toxin(s) from stool specimens. *J Clin Microbiol* 1994;32(5):1142-7.
29. Gerding DN, Johnson S, Peterson LR, Mulligan ME, Silva J Jr. *Clostridium difficile*–associated diarrhea and colitis. *Infect Control Hosp Epidemiol* 1995;16(8):459-77.
30. Kuijper EJ, de Weerdt J, Kato H, Kato N, van Dam AP, van der Vorm ER, et al. Nosocomial outbreak of *Clostridium difficile*–associated diarrhoea due to a clindamycin-resistant enterotoxin A–negative strain. *Eur J Clin Microbiol Infect Dis* 2001;20(8):528-34.
31. Olson MM, Shanholtzer CJ, Lee JT Jr, Gerding DN. Ten years of prospective *Clostridium difficile*–associated disease surveillance and treatment at the Minneapolis VA Medical Center, 1982-1991. *Infect Control Hosp Epidemiol* 1994;15(6):371-81.
32. Teasley DG, Gerding DN, Olson MM, Peterson LR, Gebhard RL, Schwartz MJ, et al. Prospective randomised trial of metronidazole versus vancomycin for *Clostridium-difficile*–associated diarrhoea and colitis. *Lancet* 1983;2(8358):1043-6.
33. Wenisch C, Parschalk B, Hasenhundl M, Hirschl AM, Graninger W. Comparison of vancomycin, teicoplanin, metronidazole, and fusidic acid for the treatment of *Clostridium difficile*–associated diarrhea. *Clin Infect Dis* 1996;22(5):813-8.
34. Young GP, Ward PB, Bayley N, Gordon D, Higgins G, Trapani JA, et al. Antibiotic-associated colitis due to *Clostridium difficile*: double-blind comparison of vancomycin with bacitracin. *Gastroenterology* 1985;89(5):1038-45.
35. De Lalla F, Nicolin R, Rinaldi E, Scarpellini P, Rigoli R, Manfrin V, et al. Prospective study of oral teicoplanin versus oral vancomycin for therapy of pseudomembranous colitis and *Clostridium difficile*–associated diarrhea. *Antimicrob Agents Chemother* 1992;36(10):2192-6.
36. Fekety R, Silva J, Kauffman C, Buggy B, Deery HG. Treatment of antibiotic-associated *Clostridium difficile* colitis with oral vancomycin: comparison of two dosage regimens. *Am J Med* 1989;86(1):15-9.
37. ASHP therapeutic position statement on the preferential use of metronidazole for the treatment of *Clostridium difficile*–associated disease. *Am J Health Syst Pharm* 1998;55(13):1407-11.
38. Dudley MN, McLaughlin JC, Carrington G, Frick J, Nightingale CH, Quintiliani R. Oral bacitracin vs vancomycin therapy for *Clostridium difficile*–induced diarrhea. A randomized double-blind trial. *Arch Intern Med* 1986;146(6):1101-4.
39. Cronberg S, Castor B, Thoren A. Fusidic acid for the treatment of antibiotic-associated colitis induced by *Clostridium difficile*. *Infection* 1984;12(4):276-9.
40. Fekety R, McFarland LV, Surawicz CM, Greenberg RN, Elmer GW, Mulligan ME. Recurrent *Clostridium difficile* diarrhea: characteristics of and risk factors for patients enrolled in a prospective, randomized, double-blinded trial. *Clin Infect Dis* 1997;24(3):324-33.
41. Bolton RP, Culshaw MA. Faecal metronidazole concentrations during oral and intravenous therapy for antibiotic associated colitis due to *Clostridium difficile*. *Gut* 1986;27(10):1169-72.
42. Tedesco FJ, Gordon D, Fortson WC. Approach to patients with multiple relapses of antibiotic-associated pseudomembranous colitis. *Am J Gastroenterol* 1985;80(11):867-8.
43. McFarland LV, Elmer GW, Surawicz CM. Breaking the cycle: treatment strategies for 163 cases of recurrent *Clostridium difficile* disease. *Am J Gastroenterol* 2002;97(7):1769-75.
44. Buggy BP, Fekety R, Silva J Jr. Therapy of relapsing *Clostridium difficile*–associated diarrhea and colitis with the combination of vancomycin and rifampin. *J Clin Gastroenterol* 1987;9(2):155-9.
45. McFarland LV, Surawicz CM, Greenberg RN, Fekety R, Elmer GW, Moyer KA, et al. A randomized placebo-controlled trial of *Saccharomyces boulardii* in combination with standard antibiotics for *Clostridium difficile* disease. *JAMA* 1994;271(24):1913-8.
46. Pochapin M. The effect of probiotics on *Clostridium difficile* diarrhea. *Am J Gastroenterol* 2000;95(1 Suppl):S11-3.
47. Tvede M, Rask-Madsen J. Bacteriotherapy for chronic relapsing *Clostridium difficile* diarrhoea in six patients. *Lancet* 1989;1(8648):1156-60.