Antimicrobial-Associated Risk Factors for *Clostridium difficile* Infection

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Antimicrobial therapy plays a central role in the pathogenesis of *Clostridium difficile* infection (CDI), presumably through disruption of indigenous intestinal microflora, thereby allowing *C. difficile* to grow and produce toxin. Investigations involving animal models and studies performed in vitro suggest that inhibitory activity against *C. difficile* and differences in the propensity to stimulate toxin production may also influence the likelihood that particular drugs may cause CDI. Although nearly all antimicrobial classes have been associated with CDI, clindamycin, third-generation cephalosporins, and penicillins have traditionally been considered to harbor the greatest risk. Recent studies have also implicated fluoroquinolones as high-risk agents, a finding that is most likely to be related in part to increasing fluoroquinolone resistance among epidemic strains (i.e., restriction-endonuclease analysis group BI/North American PFGE type 1 strains) and some nonepidemic strains of *C. difficile*. Restrictions in the use of clindamycin and third-generation cephalosporins have been associated with reductions in CDI. Because use of any antimicrobial has the potential to induce the onset of CDI and disease caused by other health care–associated pathogens, antimicrobial stewardship programs that promote judicious use of antimicrobials are encouraged in concert with environmental and infection control–related efforts.

Antimicrobial therapy plays a central role in the pathogenesis of *Clostridium difficile* infection (CDI). The presumed mechanism by which antimicrobials induce CDI is through disruption of the indigenous microflora of the colon, thereby allowing *C. difficile* to grow to high concentrations. Although nearly all classes of antimicrobials have been associated with CDI, clindamycin, third-generation cephalosporins, and penicillins have traditionally been considered to pose the greatest risk. Several recent studies have also implicated fluoroquinolones as high-risk agents. This article will review general concepts regarding the impact of antimicrobial use on *C. difficile* colonization and infection and will evaluate the risk associated with select antimicrobial agents and classes.

ANTIMICROBIAL–ASSOCIATED RISK FACTORS: GENERAL CONSIDERATIONS

Ecologic and epidemiologic characteristics of antimicrobial-associated CDI. In healthy adults, the colon contains as many as 10^{12} bacteria per gram of contents, with obligate anaerobes outnumbering facultative organisms by ~1000:1 [1]. The indigenous microflora of the colon provide an important host defense by inhibiting colonization by and overgrowth of *C. difficile* and other potential pathogens [1–3]. Antimicrobial therapy can disrupt this host defense [1]. One study suggests that, in patients with diarrhea, the diversity of the colonic microflora decreases because of overgrowth of certain types of bacteria [4]. In a series of molecular phylogenetic analyses, Young et al. [5] reported the first direct evidence of changes in the bacterial population in stool samples from a patient without CDI who had

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antimicrobial-associated diarrhea. They found that antimicrobial use caused decreases in bacterial diversity and the prevalence of butyrate-producing organisms. Most of these changes resolved within 2 weeks after the cessation of therapy.

The disruption of the indigenous flora by antimicrobials may increase the risk of CDI during therapy and for the days to weeks required for the intestinal flora to return to normal levels. Alternatively, antimicrobials that are active against *C. difficile* decrease the risk of colonization and infection during their use [1, 6]. These effects interact to create 3 broad categories by which antimicrobials affect CDI risk. Figure 1*A* and 1*B* shows data from mouse models that illustrate these 3 categories [7]. Certain agents, such as ceftriaxone, that disrupt the intestinal flora and lack significant activity against *C. difficile* promoted CDI during treatment and during the period of microflora recovery (figure 1*A* and 1*B*). Antimicrobials with inhibitory activity against *C. difficile* strains (e.g., oral vancomycin and piperacillin-tazobactam) may prevent colonization during therapy; however, such agents may facilitate colonization if exposure occurs during the period of microflora recovery [7–9]. Finally, antimicrobials that cause minimal disruption of the anaerobic microflora (e.g., aztreonam, a monobactam antimicrobial with no appreciable in vitro activity against anaerobes) did not promote CDI in mice or hamsters (figure 1*B*) [7, 10]. However, many agents that cause relatively minor disruption of the anaerobic microflora (e.g., trimethoprim-sulfamethoxazole and ciprofloxacin) have been associated with CDI [2, 11–15].

Recent observations from clinical studies suggest that antimicrobial resistance in *C. difficile* strains may be playing an increasingly important role in the epidemiology of CDI [6]. Clindamycin-resistant strains of *C. difficile* have been associated with large outbreaks of CDI [16, 17]. Clindamycin-resistant strains of *C. difficile* may thrive in an environment where other commensal flora are suppressed in the presence of clindamycin. The same concept is likely to be true for cephalosporins and fluoroquinolones when they are administered to a patient that is exposed to *C. difficile* strains that are resistant to the respective antimicrobials. Similarly, the emergence of high-level fluoroquinolone resistance among epidemic (restriction-endonuclease analysis group BI/North American PFGE type 1 [BI/NAP1]) and some nonepidemic *C. difficile* isolates has contributed to the increase in reports of an association between use of these agents and CDI due to fluoroquinolone-resistant strains [18]. Table 1 lists the general activity of a wide variety of antimicrobial agents against strains of *C. difficile* isolated over the last 2 decades, including BI/NAP1 strains [20]. Finally, associations between CDI and β-lactam/β-lactamase inhibitors, such as piperacillin-tazobactam, may be relatively infrequent, possibly because these drugs inhibit the activity of many *C. difficile* strains (i.e., *C. difficile* may be inhibited during the course of treatment with piperacillin-tazobactam) [7]. Additionally, it has also been proposed that agents such as piperacillin-tazobactam and tigecycline may be infrequently associated with CDI because they stimulate less toxin production than cefotaxime [27, 28].

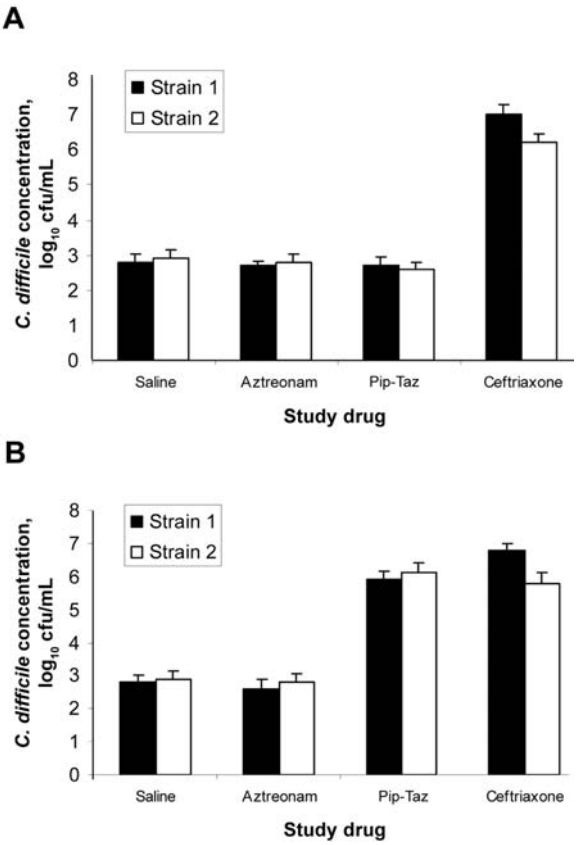

Figure 1. Effect of select agents on growth of 2 *Clostridium difficile* strains in the cecal contents of mice. Cecal contents were collected and inoculated with 10^4 cfu/mL of the *C. difficile* strains either 2 h *(A)* or 3 days *(B)* after receiving the final dose of the study drug. Samples were incubated anaerobically for 48 h, and serial dilutions were plated onto selective media for quantification of *C. difficile*. MICs were as follows: aztreonam, >128 μg/mL for both strains; piperacillin-tazobactam (Pip-Taz), 1 μg/mL for strain 1 and 2 μg/mL for strain 2; and ceftriaxone, 64 μg/mL for both strains. *Error bars,* SDs. Adapted with permission from the following article published by the American Society for Microbiology: Pultz NJ, Donskey CJ. Effect of antibiotic treatment on growth of and toxin production by *Clostridium difficile* in the cecal contents of mice. Antimicrob Agents Chemother 2005; 49:3529–32.

Not all patients who receive antimicrobials and are exposed to *C. difficile* develop CDI. This is in part attributable to other variables in the complex pathogenesis of this disease, which include the ability of the immune system to mount a serum IgG antitoxin A antibody response to *C. difficile* (figure 2) [29]. In one study, patients who did not develop increased serum anti-toxin A IgG titers in response to their first episode of CDI were 48 times as likely to develop recurrent CDI than patients who mounted an adequate immune response [30]. This is one

Table 1. Antimicrobial activity against *Clostridium difficile* strains.

Antimicrobial, by activity against *C. difficile*	MIC_{50}	MIC_{90}	Reference
Good activity			
Ampicillin	2.0	2.0	Clabots et al. [19]
Doripenem	1.0	2.0	Hecht et al. [20]
OPT-80	0.125	0.125	Hecht et al. [20]
Linezolid	0.5	2.0	Pelaez et al. [21]
Metronidazole	0.125	0.25	Hecht et al. [20]
Meropenem	2.0	2.0	Hecht et al. [20]
Nitazoxanide	0.06	0.125	Hecht et al. [20]
Penicillin G	…	1.0	Dzink and Bartlett [22]
Piperacillin	…	16	Pankuch et al. [23]
Piperacillin-tazobactam	4	4	Nord [24]
Ramoplanin	0.25	0.5	Hecht et al. [20]
Rifalazil[a]	0.0075	0.03	Hecht et al. [20]
Rifaximin[a]	0.0075	0.015	Hecht et al. [20]
Tigecycline	0.125	0.25	Hecht et al. [20]
Tinidazole	0.125	0.25	Hecht et al. [20]
Tizoxanide	0.06	0.125	Hecht et al. [20]
Vancomycin	1.0	1.0	Hecht et al. [20]
Moderate or variable activity[b]			
Clindamycin	4	>128	Clabots et al. [19]
Erythromycin	<1	>128	Clabots et al. [19]
Gatifloxacin[c]	1.0	16	Hecht et al. [20]
Moxifloxacin[c]	1.0	16	Hecht et al. [20]
Tetracycline	<1	32	Clabots et al. [19]
Poor activity			
Cefotaxime	⩾128	…	Ensminger et al. [25]
Cefoxitin	>64	>64	Pankuch et al. [23]
Cefuroxime	⩾128	…	Ensminger et al. [25]
Ciprofloxacin	8	32	Wilcox et al. [26]
Levofloxacin[c]	4	32	Hecht et al. [20]
TMP-SMZ	⩾128	⩾128	Ensminger et al. [25]

NOTE. Bold text indicates that the antimicrobial is used for investigational or approved treatment of *C. difficile* infection. One should be cautious in interpreting these data, because there is no single study that compares all of these antibiotics directly against the same collection of *C. difficile* strains. The study by Hecht et al. [20] is the most recent direct comparison. TMP-SMZ, trimethoprim-sulfamethoxazole.

[a] The specified MICs were the lowest tested. However, 3% of isolates were noted to possess high-level resistance (defined as an MIC of >256). Rifampin has similarly low MICs (data not shown).

[b] It is difficult to classify these agents on the basis of activity against *C. difficile:* although MICs of these agents were very high for some strains, MICs for most of the strains tested were very low.

[c] Higher MICs (i.e., 32–64 μg/mL) have been reported recently for these antimicrobials, although the $MICs_{50}$ and $MICs_{90}$ listed here suggest that these agents pose a moderate risk for *C. difficile* infection. Because the MICs are elevated for the recent epidemic strains (i.e., restriction-endonuclease analysis group BI/North American PFGE type 1 strains), when these agents are used in a widespread manner, they are likely to pose a risk for *C. difficile* infection that is similar to that of higher-risk agents (i.e., antimicrobials with higher MICs).

reason why elderly persons are more likely to be susceptible to CDI; other reasons include increased frequencies of hospitalization, exposure to long-term care facilities, and antimicrobial use. Some of these general risk factors are discussed in the sections that follow.

An emerging topic of interest is the increasingly frequent observations that CDI appears to be occurring more frequently in community dwelling individuals without documented or obvious traditional risk factors for CDI [31–36]. In some respects, this upsurge of patients with community-associated CDI, traditionally a health care facility–associated infection, may end up following a pattern similar to that of community-acquired methicillin-resistant *Staphylococcus aureus* (MRSA) infection. It is interesting that a tangible number of patients with community-associated CDI do not appear to have been exposed to antimicrobial agents. One characteristic that these patients seem to share is that they are more likely to have received proton-pump inhibitors (PPIs) [34, 37, 38]. For instance, a

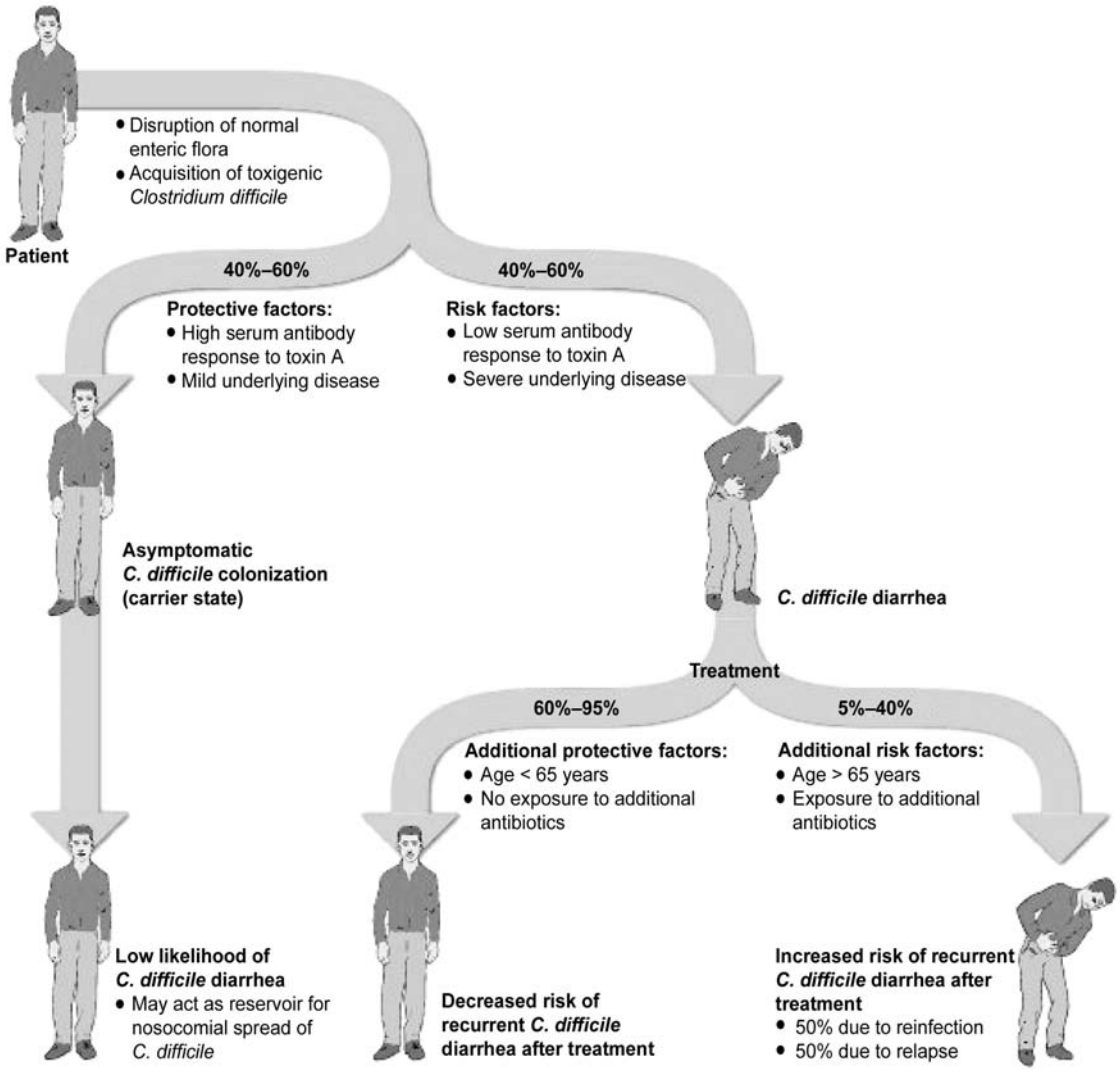

Figure 2. Factors contributing to the development of *Clostridium difficile* colonization and diarrhea following acquisition of a toxigenic strain. Adapted with permission from the following article published by the Canadian Medical Association: Poutanen SM, Simor AE. *Clostridium difficile*–associated diarrhea in adults. CMAJ 2004; 171:51–8.

case-control study based on pharmacy records in the United Kingdom found the adjusted relative risks for community-acquired CDI to be 3.5 (95% CI, 2.3–5.2) for PPI use (vs. no PPI use) and 8.2 (95% CI, 6.1–11.0) for antimicrobial use (vs. no antimicrobial use) [38]. Other studies involving larger populations also demonstrated that PPI use is a risk factor for the development of CDI [12, 39, 40], but some investigators do not agree with this finding [41].

In general, most patients with CDI were exposed to antimicrobials several weeks to several months before diagnosis [6, 11, 33, 42]. A retrospective chart review of 1364 patients for whom CDI was diagnosed found that two-thirds had received a cephalosporin during the 2-month period before diagnosis [33]. Similarly, a case-control study of hospitalized patients found that a significantly greater percentage of patients with CDI had been exposed to antimicrobials $\leqslant$6 weeks prior to diagnosis, compared with matched controls exposed $\leqslant$6 weeks before discharge (94.1% vs. 67.1%; $P < .001$) [11]. Although antimicrobial exposure remains a key risk factor for developing CDI, the presence of CDI among patients without documented exposure to antimicrobials is of concern. Some possible explanations for these cases of CDI include the enhanced virulence of the BI/NAP1 strain and increased use of PPIs. In addition, it is possible that designs of previous studies were such that patients lacking a past history of antimicrobial exposure were excluded from risk factor studies, resulting in a possible exposure bias. Nonetheless, this is an area of emerging interest that needs to be studied further.

As mentioned previously, it has been suggested that differences in the anaerobic activity of antimicrobials may account for differences in the risk of CDI associated with their use, because agents with substantial anaerobic activity cause more disruption of the colonic microflora if they are excreted into the intestinal tract. In addition, agents that disrupt the anaerobic component of the microflora without inhibiting Enterobacteriaceae or enterococci (e.g., clindamycin) may be potent promoters of *C. difficile* in the colon [1–3, 7]. However, evidence from clinical studies does not support this theory. Many agents that cause relatively minor disruption of the anaerobic microflora (e.g., fluoroquinolones) have been associated with CDI [2, 3, 11–15]. Third-generation cephalosporins, such as ceftriaxone, that are excreted in high concentrations in bile and cause extensive disruption of the anaerobic microflora have been associated with CDI, but cefotaxime, which is excreted in much lower levels in bile and causes only modest changes in the anaerobic microflora, has also been strongly associated with CDI [43].

Piperacillin-tazobactam has potent in vitro activity against anaerobes and has been independently associated with CDI in some studies [13, 44]. However, it appears to be less strongly associated with CDI than third-generation cephalosporins, possibly because β-lactam/β-lactamase inhibitors are highly active against many *C. difficile* strains [45, 46].

Concomitant use of multiple antimicrobials. Use of multiple antimicrobials has been associated with an increased risk of CDI [11, 12, 42, 47–49]. For example, a seminal case-control study found that patients who developed CDI were more likely than matched controls to have received multiple antimicrobials (80% vs. 56%; $P < .002$) [42]. In another case-control study, use of multiple antimicrobials was a significant risk factor for developing CDI (mean number used, 4.2 vs. 1.4 antimicrobials; $P < .001$) [47]. A retrospective cohort study also found that the incidence of CDI increased with the number of antimicrobials administered (relative risk, 2.01; 95% CI, 1.67–2.40) [48].

In studies that evaluate an individual antimicrobial's risk for CDI, receipt of multiple antimicrobials can lead to confounding results, making determination of risk inherently more difficult [50]. However, because many patients concurrently receive multiple antimicrobials, selection of an appropriate study design is imperative for meaningful analysis. For example, macrolide monotherapy is uncommon among hospitalized patients, and virtually all patients receiving a macrolide concurrently receive a cephalosporin for the empirical treatment of community-acquired pneumonia. Therefore, it is difficult to assess the independent contribution of each antimicrobial to the risk of developing CDI.

Duration of antimicrobial use. Prolonged antimicrobial therapy has been associated with an increased risk of CDI [13, 31, 51, 52]. The difficulties with these studies are numerous: different study designs were used, some had low numbers of cases, one was a randomized controlled trial, and most used different end points (i.e., one study used culture positivity as the sole marker for *C. difficile* [which does not equate to disease], whereas others used definitions of actual disease [toxin test positivity plus signs and symptoms of CDI]). Therefore, this literature is somewhat confusing. A retrospective cohort study of 293 hospitalized patients with CDI found an association between extended use of antimicrobial therapy and increased risk of CDI, even after adjustment for other risk factors (table 2) [13]. Cefoxitin use was the exception: longer durations of cefoxitin therapy did not demonstrate greater risk for CDI, although most patients received this antimicrobial in a single dose for short-duration preoperative prophylaxis [13]. A recent prospective, randomized clinical study of preoperative prophylaxis for colorectal surgery permitted a risk assessment for CDI associated with prophylactic antimicrobial use in this era of the circulating BI/NAP1 strain [54]. In this study, ertapenem use was associated with nearly a 3-fold greater risk of CDI than cefotetan use (1.7% vs 0.6%; $P = .22$). Although the difference was not statistically significant, a definite trend toward a higher risk for CDI was observed for ertapenem use. In addition, the percentages may look small, but considering that nearly one-third of all antimicrobial use in the hospital setting involves preoperative prophylaxis, the number of exposures to these antimicrobials is substantial. In a prospective case-control study of the epidemiology of CDI, significantly more cases with CDI than controls had received antimicrobials to treat an infection (59% vs. 31%; $P < .001$) [42]. However, significantly fewer cases had received short-course antimicrobial therapy for prophylactic purposes (20% vs. 38%; $P < .01$).

When *C. difficile* is endemic, receipt of perioperative prophylactic antimicrobial therapy for <24 h increases the risk of CDI [52]. In one study, 17 (23%) of 74 surgical patients had *C. difficile*–positive stool cultures ⩽2 weeks after receiving a single preoperative prophylactic cephalosporin dose. All of these patients had negative results of preoperative cultures; patients who tested positive for *C. difficile* before receiving the antimicrobial dose were excluded from the study [55].

Overall, prolonged antimicrobial therapy influences the risk for CDI by extending the patient's window of susceptibility to subsequent CDI. This makes it even more important for clinicians to adhere to shorter durations of therapy as data emerge to support this antimicrobial stewardship strategy (e.g., for treatment of pneumonia) and for clinicians not to unnecessarily extend antibiotic treatment "just in case" to patients who have clinically responded to therapy. Again, although longer durations of therapy are associated with a greater risk of CDI, it is important to remember that even single doses of antimicrobials

Table 2. Results of studies to determine drugs associated with a risk for *Clostridium difficile* infection.

Study, agent	No. of subjects	OR[a] (95% CI)
Lai et al. [53]		
Ciprofloxacin	92 cases, 78 controls	2.29 (1.13–166)
McCusker et al. [14]		
Clindamycin	9 cases, 7 controls	Not significant
Levofloxacin, ciprofloxacin, and/or gatifloxacin[b]	22 cases, 15 controls	12.7 (2.6–61.6)
Muto et al. [12]		
Clindamycin	32 cases, 13 controls	4.8 (1.9–12.0)
Ceftriaxone	21 cases, 8 controls	5.4 (1.8–15.8)
Levofloxacin	120 cases, 83 controls	2.0 (1.2–3.3)
Any proton-pump inhibitor	78 cases, 54 controls	2.4 (1.3–4.4)
Any histamine H_2 blocker	159 cases, 141 controls	2.0 (1.1–3.5)
Pepin et al. [13][c]		
Fluoroquinolones		
Overall	1708	3.4 (2.6–4.5)
Ciprofloxacin	1153	3.74 (2.8–4.9)
Levofloxacin	368	2.52 (1.6–3.7)
Levofloxacin and ciprofloxacin	127	4.55 (2.9–7.1)
Gatifloxacin[d]	22	6.10 (2.2–16.7)
Moxifloxacin[d]	27	Not significant
Cephalosporins		
First generation	661	1.8 (1.3–2.5)
Second generation	1001	1.9 (1.4–2.5)
Third generation	581	1.6 (1.2–2.1)
Clindamycin	147	1.8 (1.1–3.0)
Any β-lactam/β-lactamase inhibitor	355	1.9 (1.4–2.6)
Loo et al. [11]		
Any cephalosporin	115 cases, 65 controls	3.8 (2.2–6.6)
Any fluoroquinolone	128 cases, 75 controls	3.9 (2.3–6.6)
Ciprofloxacin	No data	3.1 (1.8–5.4)
Gatifloxacin or moxifloxacin	No data	3.4 (1.5–7.7)
Levofloxacin[d]	No data	Not significant
Kazakova et al. [40]		
Any cephalosporin	28 cases, 21 controls	5.19 (1.61–16.77)
Any fluoroquinolone	30 cases, 20 controls	3.22 (1.03–10.99)
Any proton-pump inhibitor	19 cases, 13 controls	5.02 (1.30–19.36)

NOTE. All studies are case controlled, unless otherwise indicated.

[a] All ORs are crude except for those from the study by Pepin et al. [13], which reported adjusted ORs.

[b] Sixty percent of cases and 60% of controls received levofloxacin, 45% of cases and 27% of controls received ciprofloxacin, and 14% of cases and 20% of controls received gatifloxacin.

[c] Retrospective cohort study involving 5619 patients who had 7421 episodes of care; *C. difficile*–associated disease (i.e., *C. difficile* infection) was diagnosed in 293 patients.

[d] The lack of significance may be misleading, because fewer patients used this antibiotic relative to the use of other antibiotics in the study.

administered for surgical prophylaxis can still increase a patient's risk.

CDI RISK FOR SELECT ANTIMICROBIAL AGENTS AND CLASSES

Historically, the antimicrobials most commonly associated with CDI in well-conducted studies are clindamycin, penicillins, and cephalosporins [56]. Perhaps because of the increasing use of fluoroquinolones among both inpatients and outpatients, use of these agents has been recently implicated as a risk factor for CDI. The association of CDI with a particular antimicrobial agent may depend on several factors, including the local prevalence of high resistance to antimicrobial agents in common use and the frequency with which the antimicrobial of interest is used.

Nearly all antimicrobials have been associated with CDI, as

can be readily noted in the US Food and Drug Administration's product labeling for antimicrobials used in the United States [15]. Figure 3 shows the magnitude of CDI risk for select antimicrobials. However, the variation between the different ORs is somewhat misleading, and comparisons of ORs from this study cannot be used to distinguish differences in risk between antimicrobials [6, 11, 12, 33, 42, 57]. Findings from many published studies conflict, primarily because of the lack of prospective, randomized, controlled studies; the use of study designs (before-and-after or quasi-experimental studies) that lack appropriate statistical analyses (such as interrupted time series analysis); the failure to achieve adequate population sizes for the antimicrobials being investigated; the inadequate group selection of controls in case-control studies; and/or the failure to adjust for substantial confounding [56, 57].

Clindamycin. Clindamycin was widely used in the 1970s and 1980s and was an agent of choice for treating infection with anaerobic organisms. However, in 1977, a clindamycin-resistant, toxigenic strain of *C. difficile* was identified as the cause of clindamycin-associated colitis in hamsters [58]. Since then, a variety of CDI outbreaks have been described, some of which were reported to result from a predominant strain [11, 18, 59, 60]. One of the first outbreaks involving clindamycin-resistant *C. difficile* (clindamycin MIC, ⩾256 μg/mL) that was formally reported and investigated occurred in 1989 and was followed by 3 more severe outbreaks in the early 1990s. The predominant strain involved in these outbreaks was highly resistant to clindamycin (100% had a clindamycin MIC of ⩾256 μg/mL); the prevalence of high-level resistance was much lower for nondominant strains (15% had an MIC of ⩾256 μg/mL) [61]. Studies of these outbreaks established that clindamycin use increased the risk of CDI. This finding led to a decrease in the use of clindamycin in US hospitals, particularly those that were impacted by CDI outbreaks, which resulted in resolution of the outbreaks, diminution of the prevalence of the epidemic strains, and decreased rates of clindamycin-associated CDI [6, 16, 61]. It is unclear whether a single-formulary intervention is generalizable to other antimicrobial classes. Clindamycin's relatively unique penchant for impacting the intestinal flora over a prolonged period may increase the window of susceptibility to CDI to a time point after the antimicrobial is discontinued, especially when the predominant strain of *C. difficile* is phenotypically resistant [62, 63]. The current BI/NAP1 epidemic strains demonstrate variable susceptibility to clindamycin. This observation, combined with the fact that clindamycin is not used as commonly as other antimicrobials in adult inpatients (except for the resurgence in its use for treatment of community-acquired MRSA), may explain why clindamycin has not been shown to be at the top of the list in every risk factor ascertainment study involving antimicrobials. Clindamycin characteristically demonstrates high MICs toward a wide range of clinical isolates of *C. difficile* (table 1).

Cephalosporins. In terms of their usefulness, a number of cephalosporins were approved for use in the 1980s and 1990s in North America and quickly gained formulary acceptance. Cephalosporin exposure soon became a strong risk factor for CDI outbreaks, probably because they became "workhorse agents" on hospital formularies, owing to their perceived safety

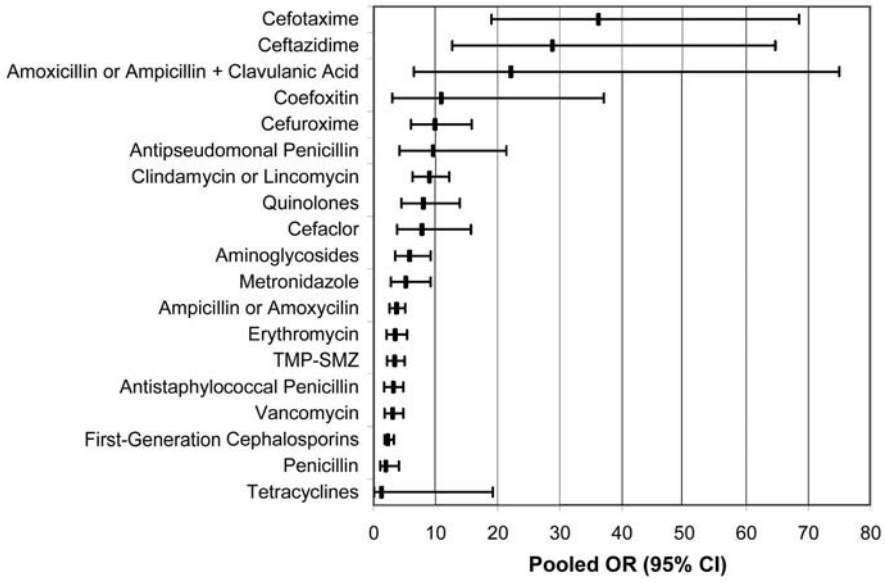

Figure 3. Meta-analysis of the risk of *Clostridium difficile* infection associated with use of select antimicrobials. TMP-SMZ, trimethoprim-sulfamethoxazole. Adapted with permission from the following article published by Elsevier: Bignardi GE. Risk factors for *Clostridium difficile* infection. J Hosp Infect 1998; 40:1–15.

and to competitive contracting [6, 15, 61]. *C. difficile* isolates are fully resistant to most cephalosporins [6, 61]. Thus, it is not surprising why cephalosporins seem to be implicated in nearly all studies to ascertain risk factors for CDI [6]. As shown in figure 3, use of second- and third-generation cephalosporins, such as cefuroxime, ceftazidime, cefotaxime, and ceftriaxone, is associated with a particularly high risk for CDI [15]. In 1994, use of second- or third-generation cephalosporins was identified as the chief risk factor for CDI in an outbreak at a Veterans Administration medical center in New York, even after controlling for use of other antimicrobial agents [49, 61]. Studies have continued to implicate cephalosporins as being strongly associated with outbreaks of CDI [11, 12]. Some studies have demonstrated that the rate of CDI was reduced after implementation of formulary interventions designed to decrease the use of cephalosporins during outbreak periods [6, 64, 65]. In the 2000s, studies continue to implicate cephalosporins as the leading antimicrobial class associated with CDI, with greater ORs than those for fluoroquinolones, despite the attention received by fluoroquinolones [11, 12, 14, 40, 66].

Fluoroquinolones. Ciprofloxacin, the first fluoroquinolone introduced in the United States, was initially considered to have a low risk for CDI. Similar to cephalosporins, fluoroquinolones became popular antimicrobials for treating inpatients and outpatients, because of their good oral bioavailability and spectra of activity. Since the introduction of ciprofloxacin, use of fluoroquinolones has been increasing in frequency and has become widespread [67], and several new agents have been introduced, including gatifloxacin, gemifloxacin, levofloxacin, moxifloxacin, and ofloxacin. As expected for a frequently used class of agents with poor in vitro activity against *C. difficile,* fluoroquinolone use has recently been associated with outbreaks of CDI (table 2) [11–14, 31, 40, 53]. The reported ORs and relative risks have ranged from 2.0 to 12.7 [12, 14]. It is difficult to compare the attributable risk of fluoroquinolone use with that of any antimicrobial use for several reasons, including variations in sample size and study design, poor selection of control subjects (some studies contained large percentages of controls who had no antimicrobial exposure), and antimicrobial polypharmacy among patients receiving all of the implicated agents [12, 57]. For example, in 2 recent studies of CDI risk factors, 32.9% and 46% of control subjects had no history of antimicrobial exposure [11, 31].

Loo et al. [11] reported an increased incidence of CDI, with relatively higher morbidity and mortality rates, caused by a predominant strain of fluoroquinolone-resistant *C. difficile* (i.e., BI/NAP1). Of the >1700 patients evaluated, 15% were randomly selected for a case-control study to identify risk factors for developing CDI. Importantly, cases were more likely than controls to acquire CDI if their duration of hospitalization was longer than that of their matched controls. In this study, cases received a significantly greater number of antimicrobials, compared with controls (1.9 vs. 1.3 antimicrobials; $P < .001$). Fluoroquinolone use and cephalosporin use were implicated as risk factors. Interestingly, clindamycin use was not implicated as a risk factor, possibly because the epidemic strain in these hospitals was clindamycin susceptible. As mentioned in table 2, levofloxacin was not significantly associated with CDI, unlike the other fluoroquinolones evaluated. Importantly, levofloxacin was not used in most of the study hospitals, and the overall number of patients exposed to levofloxacin was so small that it should not have been included in the multivariable analysis. Thus, the designation of a nonsignificant association is misleading. As discussed in the following paragraphs, several other studies have identified associations between levofloxacin and CDI [12, 13, 31].

Gaynes et al. [31] reported an outbreak of CDI that appeared to coincide with a formulary change from levofloxacin to gatifloxacin in both a long-term care facility and its adjacent hospital. During the first period of levofloxacin use (i.e., before the formulary change to gatifloxacin) in the long-term care facility, 10 of 58 patients receiving levofloxacin developed CDI [31]. The formulary change was made to gatifloxacin, and during this 9-month period, 14 of 47 patients developed CDI [31]. The formulary reverted back to levofloxacin, and an unspecified number of CDI cases were reported (the number of CDI cases was lower than the number during the period of gatifloxacin use). The ostensible outbreak that occurred during the 9-month period of gatifloxacin use involved 4 more cases of CDI than were observed during the first 9-month period of levofloxacin use. Just before the formulary switch back to levofloxacin, 10% sodium hypochlorite was used to clean the environment, and during the 9-month period of gatifloxacin use, handwashing with soap and water was reinforced. Although the reduction in the number of CDI cases was attributed to the formulary change from gatifloxacin to levofloxacin, without the use of time series analysis it is not possible to robustly conclude that the formulary change aided the CDI reduction. Additionally, the authors' conclusion ignores the contribution of the environmental cleaning done just prior to the conversion back to levofloxacin and the contribution of reinforcing handwashing with soap and water.

In a similar study involving a different patient population, rates of CDI among patients with neutropenia increased after a switch from levofloxacin to moxifloxacin prophylaxis [68]. The rate of diarrhea episodes during the initial period of levofloxacin use was 6% (10 of 159 patients), compared with 33% (42 of 132) after the switch to moxifloxacin; the rate decreased to 13% (3 of 24) after the switch back to levofloxacin. Another report also identified moxifloxacin use as a risk factor for CDI in an outbreak in a Pennsylvania hospital, which led to a formulary change from moxifloxacin back to levofloxacin [69].

However, after changing the hospital formulary from moxifloxacin back to levofloxacin, the rates of CDI actually increased further! In retrospect, there are probably 2 reasons for this. First, there is no discernible difference between fluoroquinolones (the BI/NAP1 strains are resistant to all quinolones). Second, the overall use of antimicrobials appeared to be increasing throughout all 3 periods of the study. Three studies—by Pepin et al. [13] in Quebec, Canada; Muto et al. [12] in Pittsburgh, Pennsylvania; and Kazakova et al. [40] in Augusta, Maine—demonstrated that a variety of antimicrobials, including fluoroquinolones (specifically, ciprofloxacin and levofloxacin [13]; levofloxacin, ceftriaxone, and clindamycin [12]; and levofloxacin and cephalosporins [40]), were associated with CDI. In the case-control study of 406 patients by Muto and colleagues, clindamycin (OR, 4.8; 95% CI, 1.9–12), ceftriaxone (OR, 5.4; 95% CI, 1.8–15.8), and levofloxacin (OR, 2.0; 95% CI, 1.2–3.3) were associated with CDI; the etiologic fractions for these 3 agents were 10.0%, 6.7%, and 30.8%, respectively. In fact, Kazakova and representatives from the Centers for Disease Control and Prevention indicated that outbreaks have occurred that implicate all of the currently available quinolones [40]. They suggested that the quinolones as a class should be used judiciously, because evidence now points to each quinolone as posing a risk for CDI [40]. An evaluation of elderly patients who were hospitalized for CDI that developed while they were outpatients and who did not have a history of HCF exposure in the previous 60 days demonstrated no significant differences in ORs for various fluoroquinolones (i.e., ciprofloxacin, gatifloxacin, levofloxacin, or moxifloxacin) used during the previous 30 days [70].

The emergence of *C. difficile* strains with high-level resistance to all fluoroquinolones may be contributing to outbreaks of fluoroquinolone-associated CDI. In particular, the BI/NAP1 strain has been isolated with varying frequency in at least 8 North American outbreaks since 2001, and isolates from these outbreaks have been fully resistant to all of the fluoroquinolones tested [12, 18]. A plea for HCFs to culture *C. difficile* isolates in an effort to learn more about both outbreak and nonoutbreak strains has been made [71]. Culturing the organism is not routine for any clinical laboratory in the United States, and this is part of the problem. The only way to learn more about this organism is by recovering it from culture and performing genotyping studies, susceptibility studies, and other analyses. Although additional research is needed, what has been gleaned from studies performed to date (most of which have focused on outbreaks) is that the emergence of in vitro resistance to commonly used antimicrobials and the appearance of new strains of *C. difficile* with novel virulence and survival characteristics appear to be critical factors associated with CDI outbreaks [6].

It appears that the best strategy for antimicrobial intervention is to minimize unnecessary antimicrobial exposure in a programmatic fashion, which can be accomplished through a comprehensive antimicrobial stewardship program [18, 31, 57, 72, 73]. Switching from one quinolone to another is futile, as has been demonstrated by a number of studies thus far discussed. The most likely explanation for this finding is that quinolones share equal risk in terms of their association with CDI, and nearly all of them possess high MICs for BI/NAP1 epidemic strains. It is important to reiterate that antimicrobial intervention alone is unlikely to result in successful control of a CDI outbreak, and issues related to the environment and infection control should also be addressed. Comprehensive antimicrobial stewardship efforts in conjunction with proper environmental disinfection, hand hygiene compliance, and single-room isolation or cohorting are likely to yield the largest benefit to controlling and preventing outbreaks of CDI. Examples of this "bundled" approach are discussed later in this article. Elsewhere, Muto et al. [74] and Valiquette et al. [75] have evaluated the bundled approach and its success.

ANTIMICROBIAL STEWARDSHIP EFFORTS TO PREVENT AND CONTROL CDI OUTBREAKS

Evidence from several studies suggests that changes in antimicrobial prescribing practices in hospitals can affect the incidence of HCF-acquired non-BI/NAP1 CDI [76]. Two studies described interventions in which clindamycin use was restricted as a means to control CDI outbreaks. In both studies, clindamycin use was associated with increases in the incidence of CDI. The high CDI incidence persisted despite increased use of infection control measures but decreased with the restriction of clindamycin use [16, 17]. For reasons explained previously in this article, clindamycin exposure appears to be unique, at least in animal models, resulting in a longer window of susceptibility to CDI; thus, the results from these formulary intervention studies may not be readily generalizable to other antimicrobials.

Restrictions in cephalosporin use for control of CDI have been investigated in 3 studies. One study involving an elderly care unit of a United Kingdom hospital where antimicrobial restriction and infection control policies reduced the use of cefuroxime by 90% found that the number of CDIs was reduced from 37 in the 7 months before the intervention to 16 in the 7 months after the intervention ($P < .001$) [64]. In another United Kingdom hospital, CDI rates increased following a formulary change from cefotaxime to ceftriaxone for the initial treatment of severe sepsis or pneumonia [65]. Despite infection control measures and an increasingly restrictive policy on antimicrobial use that culminated in withdrawal of all oral cephalosporin therapy, CDI persisted. Finally, the incidence of CDI decreased 6 months after the substitution of levofloxacin for ceftriaxone. The investigators speculated that the delay resulted

from long-lived environmental reservoirs of *C. difficile* that persisted until several months after the introduction of levofloxacin. The *C. difficile* isolates implicated in this outbreak were not characterized, and given the location and timing of this study, it is not likely that the BI/NAP1 fluoroquinolone-resistant strain predominated. In a US facility, a 75% increase in the rate of CDI prompted implementation of an antimicrobial management program intended to minimize the use of third-generation cephalosporins. Use of third-generation cephalosporins decreased after implementation of the policy, as did the rate of CDI ($P = .002$) [77].

An evaluation of the effect of infection control interventions and antimicrobial stewardship efforts on CDI rates was recently undertaken in Montreal [75]. The first wave of multifaceted interventions described in this study involved strict adherence to infection control policies, including use of dedicated equipment; isolation of patients who had diarrhea of unknown etiology, and retaining patients with CDI in isolation until discharge; and environmental cleaning with sodium hypochlorite, followed by 7% accelerated hydrogen peroxide for terminal disinfection of rooms occupied by patients with CDI [75]. The second wave of interventions involved the formation of an antimicrobial stewardship program facilitated by infectious diseases physicians and pharmacists. Guidelines were created, education was performed, and both were followed up by prospective audit with feedback rather than a prior authorization (i.e., restrictive) approach. Antimicrobial stewardship interventions were effective in reducing the use of antimicrobials that were highly associated with CDI (e.g., cephalosporins, most fluoroquinolones, and macrolides), as well as reducing overall antimicrobial use. The combined approach of infection control with environmental and antimicrobial stewardship significantly resulted in reduced rates of CDI. Importantly, this was accomplished without the implementation of antimicrobial restrictions. Rather, another stewardship technique—prospective audit with feedback—demonstrated success in reducing overall antimicrobial use and significantly diminishing rates of CDI.

Guidelines for enhancing antimicrobial use through institutional antimicrobial stewardship programs were recently published by the Infectious Diseases Society of America and the Society for Healthcare Epidemiology of America [78]. Reducing unnecessary antimicrobial use is a primary objective of these programs, and they have been shown to reduce rates of CDI. Specific elements of these programs that can be implemented by hospitals to minimize CDI include reducing the duration of antimicrobial use, reducing "redundant" antimicrobial therapy, and switching from parenteral therapy to oral therapy (to help facilitate the patient's transition from a high-risk facility to their home, where they are less likely to come into contact with *C. difficile,* as well as to shorten the duration of antimicrobial therapy). The guidelines are helpful in describing the necessary resources and the types of programs that have been shown to be effective in reducing unnecessary antimicrobial use.

As part of a *C. difficile* control protocol at the University of Pittsburgh (Pittsburgh), use of antimicrobials associated with an increased risk of CDI (i.e., clindamycin, ceftriaxone, and levofloxacin) was restricted as part of a more global antimicrobial management program [12]. This resulted in decreased use of fluoroquinolones (by 50% from preintervention use), clindamycin (by 75%), and ceftriaxone (by 35%). However, the rate of nosocomial CDI at this institution was already reduced by 50% with infection control measures alone, and the investigators believed that the antimicrobial restrictions may have contributed to the sustainability of low rates of CDI ($\leqslant$5.0 infections per 1000 discharges for the past 5 years) [12]. Methods used by the "bundled" approach at the University of Pittsburgh included education, enhanced case finding, expanded infection control measures, the formation of a *C. difficile* management team, and implementation of an antimicrobial stewardship program [74]. This bundled program, which targeted all aspects of CDI, resulted in an impressive 78% reduction in the overall rate of CDI between 2002 and 2006 [74].

Definitive evidence on the value of antimicrobial restriction alone, or of any other component of a bundled multifaceted approach, in decreasing CDI rates requires formal study of compliance with a single intervention and comparison of its outcomes (i.e., nosocomial CDI rates) to those for a control population treated identically, apart from implementation of the intervention under study. This may not be feasible if patient safety is a concern, but it is certainly possible for testing new interventions when proven standard interventions are already in place. A CDI rate as close as possible to 0 should be the goal by implementing, at minimum, interventions that have been demonstrated in controlled trials to be beneficial [74]. It is important to realize that if there is no exposure to *C. difficile,* CDI cannot occur regardless of the antimicrobials used. It is equally important to understand that the corollary position is also true: if no antimicrobials are used, *C. difficile* exposure will likely result in fewer cases of CDI. Optimal results are likely to occur through good antimicrobial stewardship efforts combined with infection control best practices that involve environmental cleaning. One of the primary drivers for health care facilities investing in antimicrobial stewardship programs, as well as in infection control and robust environmental services, is to minimize their patients' probability of developing CDI during or following hospitalization. The articles by Valiquette et al. [75] and Muto et al. [74] published this past year demonstrate the vital and successful synergistic interrelationship between these 3 critical components (i.e., formalized antimicrobial stewardship programs, infection control programs, and environmental services departments) for any health care facility

in an era when CDI has been proven to be a formidable and lethal infectious disease.

CONCLUSION

Disruption of colonization resistance during antimicrobial use helps create conditions favorable to *C. difficile* proliferation after *C. difficile* is ingested. In turn, patients who develop CDI are likely to contaminate their environment, which increases the probability of transmission, particularly to persons with an increased risk of *C. difficile* colonization (secondary to antimicrobial exposure). Use of any antimicrobial can be a risk factor for intestinal colonization with *C. difficile*.

Although CDI can occur as a result of exposure to any antimicrobial, it has most been associated with use of clindamycin, cephalosporins, and fluoroquinolones. The latter 2 classes currently remain in common use in HCFs. In many cases, CDI outbreaks are multifactorial. CDI rates often strongly correlate with increasing total antimicrobial consumption, introduction of a particular strain of *C. difficile*, poor attention to environmental cleaning, and waning compliance with good infection control practices. Because CDI outbreaks are often multifactorial in terms of cause, the prevention and control of outbreaks must involve multiple interventions aimed at improving antimicrobial use across the health care system and in individual patients, as well as interventions targeting environment control, personnel hand hygiene, and barrier precautions, which are discussed in another article in this supplement [79].

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References

1. Donskey CJ. The role of the intestinal tract as a reservoir and source for transmission of nosocomial pathogens. Clin Infect Dis **2004**; 39: 219–26.
2. Vollaard EJ, Clasener HA. Colonization resistance. Antimicrob Agents Chemother **1994**; 38:409–14.
3. Wilson KH. The microecology of *Clostridium difficile*. Clin Infect Dis **1993**; 16(Suppl 4):S214–8.
4. Mai V, Braden CR, Heckendorf J, Pironis B, Hirshon JM. Monitoring of stool microbiota in subjects with diarrhea indicates distortions in composition. J Clin Microbiol **2006**; 44:4550–2.
5. Young VB, Schmidt TM. Antibiotic-associated diarrhea accompanied by large-scale alterations in the composition of the fecal microbiota. J Clin Microbiol **2004**; 42:1203–6.
6. Gerding DN. Clindamycin, cephalosporins, fluoroquinolones, and *Clostridium difficile*–associated diarrhea: this is an antimicrobial resistance problem. Clin Infect Dis **2004**; 38:646–8.
7. Pultz NJ, Donskey CJ. Effect of antibiotic treatment on growth of and toxin production by *Clostridium difficile* in the cecal contents of mice. Antimicrob Agents Chemother **2005**; 49:3529–32.
8. Fekety R, Silva J, Toshniwal R, et al. Antibiotic-associated colitis: effects of antibiotics on *Clostridium difficile* and the disease in hamsters. Rev Infect Dis **1979**; 1:386–97.
9. Rolfe RD, Finegold SM. Intestinal beta-lactamase activity in ampicillin-induced, *Clostridium difficile*–associated ileocecitis. J Infect Dis **1983**; 147:227–35.
10. Borriello SP, Barclay FE, Welch AR. Evaluation of the predictive capability of an in-vitro model of colonization resistance to *Clostridium difficile* infection. Microb Ecol Health Dis **1988**; 1:61–4.
11. Loo VG, Poirier L, Miller MA, et al. A predominantly clonal multi-institutional outbreak of *Clostridium difficile*–associated diarrhea with high morbidity and mortality. N Engl J Med **2005**; 353:2442–9.
12. Muto CA, Pokrywka M, Shutt K, et al. A large outbreak of *Clostridium difficile*–associated disease with an unexpected proportion of deaths and colectomies at a teaching hospital following increased fluoroquinolone use. Infect Control Hosp Epidemiol **2005**; 26:273–80.
13. Pepin J, Saheb N, Coulombe MA, et al. Emergence of fluoroquinolones as the predominant risk factor for *Clostridium difficile*–associated diarrhea: a cohort study during an epidemic in Quebec. Clin Infect Dis **2005**; 41:1254–60.
14. McCusker ME, Harris AD, Perencevich E, Roghmann MC. Fluoroquinolone use and *Clostridium difficile*–associated diarrhea. Emerg Infect Dis **2003**; 9:730–3.
15. Bignardi GE. Risk factors for *Clostridium difficile* infection. J Hosp Infect **1998**; 40:1–15.
16. Pear SM, Williamson TH, Bettin KM, Gerding DN, Galgiani JN. Decrease in nosocomial *Clostridium difficile*–associated diarrhea by restricting clindamycin use. Ann Intern Med **1994**; 120:272–7.
17. Climo MW, Israel DS, Wong ES, Williams D, Coudron P, Markowitz SM. Hospital-wide restriction of clindamycin: effect on the incidence of *Clostridium difficile*–associated diarrhea and cost. Ann Intern Med **1998**; 128(12 pt 1):989–95.
18. McDonald LC, Killgore GE, Thompson A, et al. An epidemic, toxin gene-variant strain of *Clostridium difficile*. N Engl J Med **2005**; 353: 2433–41.
19. Clabots CR, Shanholtzer CJ, Peterson LR, Gerding DN. In vitro activity of efrotomycin, ciprofloxacin, and six other antimicrobials against *Clostridium difficile*. Diagn Microbiol Infect Dis **1987**; 6:49–52.
20. Hecht DW, Galang MA, Sambol SP, Osmolski JR, Johnson S, Gerding DN. In vitro activities of 15 antimicrobial agents against 110 toxigenic *Clostridium difficile* clinical isolates collected from 1983 to 2004. Antimicrob Agents Chemother **2007**; 51:2716–9.
21. Pelaez T, Alonso R, Perez C, Alcala L, Cuevas O, Bouza E. In vitro activity of linezolid against *Clostridium difficile*. Antimicrob Agents Chemother **2002**;46:1617–8.
22. Dzink J, Bartlett JG. In vitro susceptibility of *Clostridium difficile* isolates from patients with antibiotic-associated diarrhea or colitis. Antimicrob Agents Chemother 1980; 17:695–8.
23. Pankuch GA, Jacobs MR, Appelbaum PC. Susceptibilities of 428 gram-positive and -negative anaerobic bacteria to Bay y3118 compared with their susceptibilities to ciprofloxacin, clindamycin, metronidazole, piperacillin, piperacillin−tazobactam, and cefoxitin. Antimicrob Agents Chemother **1993**; 37:1649–54.
24. Nord CE. In vitro activity of quinolones and other antimicrobial agents against anaerobic bacteria. Clin Infect Dis **1996**; 23(Suppl 1):S15–8.
25. Ensminger PW, Counter FT, Thomas LJ, Lubbehusen PP. Susceptibility,

resistance development, and synergy of antimicrobial combinations against *Clostridium difficile*. Curr Microbiol **1982**; 7:59–62.
26. Wilcox MH, Fawley W, Freeman J, Brayson J. In vitro activity of new generation fluoroquinolones against genotypically distinct and indistinguishable *Clostridium difficile* isolates. J Antimicrob Chemother **2000**; 46:551–6.
27. Baines SD, Freeman J, Wilcox MH. Effects of piperacillin/tazobactam on *Clostridium difficile* growth and toxin production in a human gut model. J Antimicrob Chemother **2005**; 55:974–82.
28. Freeman J, Wilcox MH. Ureidopenicillins and risk of *Clostridium difficile* infection. J Antimicrob Chemother **2001**; 47:719.
29. Poutanen SM, Simor AE. *Clostridium difficile*–associated diarrhea in adults. CMAJ **2004**; 171:51–8.
30. Kyne L, Warny M, Qamar A, Kelly CP. Association between antibody response to toxin A and protection against recurrent *Clostridium difficile* diarrhoea. Lancet **2001**; 357:189–93.
31. Gaynes R, Rimland D, Killum E, et al. Outbreak of *Clostridium difficile* infection in a long-term care facility: association with gatifloxacin use. Clin Infect Dis **2004**; 38:640–5.
32. Johal SS, Hammond J, Solomon K, James PD, Mahida YR. *Clostridium difficile*–associated diarrhoea in hospitalised patients: onset in the community and hospital and role of flexible sigmoidoscopy. Gut **2004**; 53: 673–7.
33. Pépin J, Valiquette L, Alary ME, et al. *Clostridium difficile*–associated diarrhea in a region of Quebec from 1991 to 2003: a changing pattern of disease severity. CMAJ **2004**; 171:466–72.
34. Kutty P, Benoit S, Woods C, et al. Emerging *Clostridium difficile*–associated disease in the community and the role of non-antimicrobial risk factors. In: Program and abstracts of the 44th Annual Meeting of the infectious Diseases Society of America (Toronto, Canada). **2006**.
35. Gaynes RP, Rimland D, McDonald C. Community-acquired *Clostridium difficile*–associated diarrhea (CDAD) among outpatients in a VA medical center [poster L-1550]. In: Program and abstracts of the 46th Annual Interscience Conference on Antimicrobials and Chemotherapy (San Francisco, CA). **2006**.
36. Centers for Disease Control and Prevention. Severe *Clostridium difficile*–associated disease in populations previously at low risk—four states, 2005. MMWR Morb Mortal Wkly Rep **2005**; 54:1201–28.
37. Dial S, Delaney JA, Barkun AN, Suissa S. Use of gastric acid–suppressive agents and the risk of community-acquired *Clostridium difficile*–associated disease. JAMA **2005**; 294:2989–95.
38. Dial S, Delaney JA, Schneider V, Suissa S. Proton pump inhibitor use and risk of community-acquired *Clostridium difficile*–associated disease defined by prescription for oral vancomycin therapy. CMAJ **2006**; 175: 745–8.
39. Cunningham R, Dale B, Undy B, Gaunt N. Proton pump inhibitors as a risk factor for *Clostridium difficile* diarrhoea. J Hosp Infect **2003**;54: 243–5.
40. Kazakova SV, Ware K, Baughman B, et al. A hospital outbreak of diarrhea due to an emerging epidemic strain of *Clostridium difficile*. Arch Intern Med **2006**; 166:2518–24.
41. Lowe DO, Mamdani MM, Kopp A, Low DE, Juurlink DN. Proton pump inhibitors and hospitalization for *Clostridium difficile*–associated disease: a population-based study. Clin Infect Dis **2006**; 43:1272–6.
42. Gerding DN, Olson MM, Peterson LR, et al. *Clostridium difficile*–associated diarrhea and colitis in adults: a prospective case-controlled epidemiologic study. Arch Intern Med **1986**; 146:95–100.
43. Sullivan A, Edlund C, Nord CE. Effect of antimicrobial agents on the ecological balance of human microflora. Lancet Infect Dis **2001**; 1: 101–14.
44. Harbarth S, Samore MH, Carmeli Y. Antibiotic prophylaxis and the risk of *Clostridium difficile*–associated diarrhoea. J Hosp Infect **2001**; 48:93–7.
45. Settle CD, Wilcox MH, Fawley WN, Corrado OJ, Hawkey PM. Prospective study of the risk of *Clostridium difficile* diarrhoea in elderly patients following treatment with cefotaxime or piperacillin-tazobactam. Aliment Pharmacol Ther **1998**; 12:1217–23.
46. Wilcox MH, Freeman J, Fawley W, et al. Long-term surveillance of cefotaxime and piperacillin-tazobactam prescribing and incidence of *Clostridium difficile* diarrhoea. J Antimicrob Chemother **2004**; 54: 168–72.
47. Thibault A, Miller MA, Gaese C. Risk factors for the development of *Clostridium difficile*–associated diarrhea during a hospital outbreak. Infect Control Hosp Epidemiol **1991**; 12:345–8.
48. Chang VT, Nelson K. The role of physical proximity in nosocomial diarrhea. Clin Infect Dis **2000**; 31:717–22.
49. Nelson DE, Auerbach SB, Baltch AL, et al. Epidemic *Clostridium difficile*–associated diarrhea: role of second- and third-generation cephalosporins. Infect Control Hosp Epidemiol **1994**; 15:88–94.
50. Wilcox MH. Clarithromycin and risk of *Clostridium difficile*–associated diarrhoea. J Antimicrob Chemother **2001**; 47:358–9.
51. Zimmerman RK. Risk factors for *Clostridium difficile* cytotoxin–positive diarrhea after control for horizontal transmission. Infect Control Hosp Epidemiol **1991**; 12:96–100.
52. Yee J, Dixon CM, McLean AP, Meakins JL. *Clostridium difficile* disease in a department of surgery: the significance of prophylactic antibiotics. Arch Surg **1991**; 126:241–6.
53. Lai KK, Melvin ZS, Menard MJ, Kotilainen HR, Baker S. *Clostridium difficile*–associated diarrhea: epidemiology, risk factors, and infection control. Infect Control Hosp Epidemiol **1997**; 18:628–32.
54. Itani KM, Wilson SE, Awad SS, Jensen EH, Finn TS, Abramson MA. Ertapenem versus cefotetan prophylaxis in elective colorectal surgery. N Engl J Med **2006**; 355:2640–51.
55. Privitera G, Scarpellini P, Ortisi G, Nicastro G, Nicolin R, de Lalla F. Prospective study of *Clostridium difficile* intestinal colonization and disease following single-dose antibiotic prophylaxis in surgery. Antimicrob Agents Chemother **1991**; 35:208–10.
56. Thomas C, Stevenson M, Riley TV. Antibiotics and hospital-acquired *Clostridium difficile*–associated diarrhoea: a systematic review. J Antimicrob Chemother **2003**; 51:1339–50.
57. Owens RC. *Clostridium difficile*–associated disease: an emerging threat to patient safety: insights from the Society of Infectious Diseases Pharmacists. Pharmacotherapy **2006**; 26:299–311.
58. Bartlett JG, Onderdonk AB, Cisneros RL, Kasper DL. Clindamycin-associated colitis due to a toxin-producing species of *Clostridium* in hamsters. J Infect Dis **1977**; 136:701–5.
59. Samore MH, Bettin KM, DeGirolami PC, Clabots CR, Gerding DN, Karchmer AW. Wide diversity of *Clostridium difficile* types at a tertiary referral hospital. J Infect Dis **1994**; 170:615–21.
60. Samore M, Killgore G, Johnson S, et al. Multicenter typing comparison of sporadic and outbreak *Clostridium difficile* isolates from geographically diverse hospitals. J Infect Dis **1997**; 176:1233–8.
61. Johnson S, Samore MH, Farrow KA, et al. Epidemics of diarrhea caused by a clindamycin-resistant strain of *Clostridium difficile* in four hospitals. N Engl J Med **1999**; 341:1645–51.
62. Sambol SP, Merrigan MM, Tang JK, Johnson S, Gerding DN. Colonization for the prevention of *Clostridium difficile* disease in hamsters. J Infect Dis **2002**; 186:1781–9.
63. Larson HE, Borriello SP. Quantitative study of antibiotic-induced susceptibility to *Clostridium difficile* enterocecitis in hamsters. Antimicrob Agents Chemother **1990**; 34:1348–53.
64. McNulty C, Logan M, Donald IP, et al. Successful control of *Clostridium difficile* infection in an elderly care unit through use of a restrictive antibiotic policy. J Antimicrob Chemother **1997**; 40:707–11.
65. Khan R, Cheesbrough J. Impact of changes in antibiotic policy on *Clostridium difficile*–associated diarrhoea (CDAD) over a five-year period in a district general hospital. J Hosp Infect **2003**; 54:104–8.
66. Owens RC. *Clostridium difficile*–associated disease: changing epidemiology and implications for management. Drugs **2007**; 67:487–502.
67. Linder JA, Huang ES, Steinman MA, Gonzales R, Stafford RS. Fluoroquinolone prescribing in the United States: 1995 to 2002. Am J Med **2005**; 118:259–68.

68. von Baum H, Sigge A, Bommer M, et al. Moxifloxacin prophylaxis in neutropenic patients. J Antimicrob Chemother **2006**; 58:891–4.
69. Biller P, Shank B, Lind L, et al. Moxifloxacin therapy as a risk factor for *Clostridium difficile*–associated disease during an outbreak: attempts to control a new epidemic strain. Infect Control Hosp Epidemiol **2007**; 28:198–201.
70. Dhalla IA, Mamdani MM, Simor AE, Kopp A, Rochon PA, Juurlink DN. Are broad-spectrum fluoroquinolones more likely to cause *Clostridium difficile*–associated disease? Antimicrob Agents Chemother **2006**; 50:3216–9.
71. Gerding DN. New definitions will help, but cultures are critical for resolving unanswered questions about *Clostridium difficile*. Infect Control Hosp Epidemiol **2007**; 28:113–5.
72. Davey P, Brown E, Fenelon L, et al. Systematic review of antimicrobial drug prescribing in hospitals. Emerg Infect Dis **2006**; 12:211–6.
73. Muto CA, Jernigan JA, Ostrowsky BE, et al. SHEA guideline for preventing nosocomial transmission of multidrug-resistant strains of *Staphylococcus aureus* and enterococcus. Infect Control Hosp Epidemiol **2003**; 24:362–86.
74. Muto CA, Blank MK, Marsh JW, et al. Control of an outbreak of infection with the hypervirulent *Clostridium difficile* BI strain in a university hospital using a comprehensive "bundle" approach. Clin Infect Dis **2007**; 45:1266–73.
75. Valiquette L, Cossette B, Garant MP, Diab H, Pepin J. Impact of a reduction in the use of high-risk antibiotics on the course of an epidemic of *Clostridium difficile*–associated disease caused by the hypervirulent NAP1/027 strain. Clin Infect Dis **2007**; 45(Suppl 2):S112–21.
76. Davey P, Brown E, Fenelon L, et al. Interventions to improve antibiotic prescribing practices for hospital inpatients. Cochrane Database Syst Rev **2005**:CD003543.
77. Carling P, Fung T, Killion A, Terrin N, Barza M. Favorable impact of a multidisciplinary antibiotic management program conducted during 7 years. Infect Control Hosp Epidemiol **2003**; 24:699–706.
78. Dellit TH, Owens RC, McGowan JE Jr, et al. Infectious Diseases Society of America and the Society for Healthcare Epidemiology of America guidelines for developing an institutional program to enhance antimicrobial stewardship. Clin Infect Dis **2007**; 44:159–77.
79. Gerding DN, Muto CA, Owens RC Jr. Measures to control and prevent *Clostridium difficile* infection. Clin Infect Dis **2008**; 46(Suppl 1):S43–9 (in this supplement).