

Antibiotics and Collateral Damage

Daryl J. Hoban, PhD, FCCM, D(ABMM)

Professor of Medical Microbiology
University of Manitoba
Clinical Microbiologist
Health Sciences Centre
Winnipeg, Manitoba, Canada

The precarious stability of the normal indigenous flora of the human gastrointestinal tract may be disturbed by a number of factors, but frequently and crucially by antibiotic therapy. This article explores recent insights on the collateral and ecologic effects of antibiotics on the microbiologic flora of the human body and the possible consequences of those effects, which are just beginning to be better understood. New data on this subject will not only help in designing better clinical trials but also begin to answer key questions about collateral damage. Clinical Cornerstone® Monograph. Copyright © 2004 Excerpta Medica, Inc.

“The normal indigenous flora of the human gastrointestinal tract comprises a remarkably complex yet stable colony of more than 400 separate species, living in a symbiotic relationship with the human host.” (1) It has been said that we prescribe antimicrobials to treat bacteria rather than to treat patients. Clinicians often target specific sites, such as the respiratory tract, soft tissues, or urinary tract; particular conditions, such as bacteremia or meningitis; or individual pathogens at a particular site of infection. But antimicrobials do not “know” this and invariably have effects on body flora in sites other than where the infecting pathogen resides.

Over the past decade a broad consensus has emerged recognizing that the future of antibiotic treatment will depend on the evolution of bacterial resistance (2), not only of the pathogens targeted by therapy but also, perhaps most importantly, of commensal organisms (3). Research today clearly demonstrates that gene resistance mechanisms and transfer of resistant genes are occurring in commensal organisms through collateral effects in addition to the pathogenic organisms being targeted with antibiotics. The evaluation of the ecologic impact of antimicrobial use encompasses the emergence and spread of resistance genes and strains as well as modifications to the distribution of micro-

KEY POINT

Over the past decade a broad consensus has emerged recognizing that the future of antibiotic treatment will depend on the evolution of bacterial resistance, not only of the pathogens targeted by therapy but also, perhaps most importantly, commensal organisms.

bial populations in human commensal flora. This awareness raises new questions, which are leading to novel avenues for research (4).

EFFECTS OF ANTIMICROBIALS ON BODY FLORA

What effects do antimicrobials have on commensals in the human body? To answer this question, the effects they have on susceptible as opposed to resistant organisms must be examined. It is also necessary to understand the “bystander effects” of antimicrobials rather than their direct effects on pathogens. Broader knowledge of the effects of antimicrobials, particularly reduction of colonization resistance, may help prevent colonization with

pathogenic bacteria. Organism overgrowth as it may occur in the bowel or in the oropharynx is critical to our understanding of collateral effects. The mechanism by which susceptible clones replace resistant clones, and whether resistant clones can be transferred from patient to patient or from one facility to another, are also important.

Anaerobic bacteria are widespread bowel commensals. Select fluoroquinolones, which are widely used in treating respiratory tract infections because of their activity against respiratory organisms, are also active against commensal gut anaerobes. In addition, fluoroquinolones, as well as being very active against *Streptococcus pneumoniae*, *Haemophilus influenzae*, and *Moraxella catarrhalis*, are also highly active against Enterobacteriaceae, which are also predominant bowel flora. This attribute contrasts with other antimicrobial groups that have minimal effects against anaerobes. If anaerobes are potentially at risk from bystander effects, it is important to consider the positive benefits of anaerobes for man.

Anaerobes are crucial to colonization resistance. In the mid-1980s when gut sterilization was performed in neutropenic patients prior to chemotherapy, these patients, with an iatrogenic lack of appropriate gut flora, subsequently colonized with more vigorous pathogens and became extremely ill. As a result, disturbances of gut flora are now minimized as much as possible in patients undergoing chemotherapy. Anaerobes are also involved in nonspecific immunogenic stimulus, vitamin K production, and bile acid deconjugation. Therefore, a key question in the treatment of lower respiratory tract infections (LRTIs) is if anaerobes are benefi-

cial, why should we be targeting them directly or indirectly with antibiotics when we are really treating respiratory pathogens?

The fluoroquinolones, particularly gatifloxacin, levofloxacin, and moxifloxacin, have become formidable therapeutic options (5,6) because of their safety profile, low levels of resistance, and activity against gram-positive organisms such as *S pneumoniae*. These are advocated as joint first-choice agents for community-acquired pneumonia by expert bodies such as the Infectious Diseases Society of America (7). In contrast, because of potential bystander effects, inappropriate use (8), low activity against commensals with a potential for acquiring resistance such as *Pseudomonas* and *Bacteroides* (9), and increasing resistance (10) and cross-resistance (11), some authorities such as the American Thoracic Society (12) and the British Thoracic Society (13) have recommended restricting the use of fluoroquinolones (14) and have argued that fluoroquinolones should be reserved for special cases or as second-line agents in the treatment of LRTIs.

EFFECTS OF ANTIMICROBIALS ON GUT FLORA

Table I shows the activity in terms of minimum inhibitory concentrations for 90% of strains (MIC_{90}) of fluoroquinolones against selected bowel organisms (15). Overall these antimicrobials exhibit activity against *Escherichia coli* and *Klebsiella pneumoniae* but lesser activity against *Pseudomonas aeruginosa* and *Bacteroides fragilis*, and individual agents have varying in vitro activity. The questions of whether and how that equates to

TABLE I. IN VITRO ACTIVITY OF FLUOROQUINOLONES (GRAM-NEGATIVE BACILLI)

Pathogens	MIC_{90} ($\mu\text{g/mL}$)			
	Ciprofloxacin	Gatifloxacin	Levofloxacin	Moxifloxacin
<i>Escherichia coli</i>	0.12	0.1	0.12	0.5
<i>Klebsiella pneumoniae</i>	0.06	0.1	0.25	0.5
<i>Pseudomonas aeruginosa</i>	4	16	8	32
<i>Bacteroides fragilis</i>	16	1	8	1

MIC_{90} = minimum inhibitory concentration required to inhibit 90% of isolates. Adapted with permission (15).

the effect these agents have in the gut are more problematic.

Antibiotics also vary in their activity against anaerobes such as *B fragilis*. **Table II** shows MIC₅₀ and MIC₉₀ not just for quinolones but for other typical anaerobic agents such as clindamycin, metronidazole, and amoxicillin/clavulanate (16). All these agents show an extreme range of activity, with ciprofloxacin and levofloxacin the lowest. When examining anaerobes other than *B fragilis* in relation to fluoroquinolones, a similar pattern of activity is noted (**Table III**) (15). *Clostridium difficile* is particularly interesting when viewed from the ecologic impact of antibiotic therapy. Despite the wide range of activities, it can be seen in these tables that the respiratory fluoroquinolones, gatifloxacin, moxifloxacin, and trovafloxacin, in particular, have enhanced in vitro activity against anaerobes compared with ciprofloxacin or levofloxacin.

In our laboratory at the Health Sciences Center, we are undertaking investigational work on the effect of antimicrobials on bowel organisms, not just by examining MICs but also by using kill curves and a colon model to explore the interaction of these agents with potential pathogens that are indirectly being targeted in the bowel. **Figure 1** demonstrates a kill curve of *B fragilis* using 4 times the MIC of a number of antibiotics (16). Despite the varying MICs of these drugs and whether or not they are considered to have enhanced anaerobe activity, given a sufficient concentration of drug as a multiple of its MIC, within 24 hours all of these drugs will kill the *B fragilis* strain. Thus, at high concentrations, even drugs not considered to have intrinsic anaerobe activity can kill some anaerobic bacteria.

FLUOROQUINOLONES AND COLONIC ANAEROBES (*B FRAGILIS*): UNANSWERED QUESTIONS

In considering the advantages and disadvantages of

TABLE II. ANTIMICROBIAL ACTIVITY AGAINST 101 BACTEROIDES FRAGILIS (16)

	MIC (µg/mL)							
	Amox/Clav	Cipro	Clinda	Gati	Gemi	Levo	Metro	Moxi
MIC ₅₀	1	16	2	1	1	8	1	1
MIC ₉₀	4	64	4	4	16	32	2	4
Range	0.125–64	2–512	0.03–32	0.25–64	0.12–128	1–512	0.12–4	0.12–32

MIC = minimum inhibitory concentration; Amox/Clav = amoxicillin/clavulanate; Cipro = ciprofloxacin; Clinda = clindamycin; Gati = gatifloxacin; Gemi = gemifloxacin; Levo = levofloxacin; Metro = metronidazole; Moxi = moxifloxacin.

TABLE III. IN VITRO ACTIVITY OF THE NEW FLUOROQUINOLONES AGAINST ANAEROBES

Pathogens	MIC ₉₀ (µg/mL)					
	Cipro	Gati	Gemi	Levo	Moxi	Trova
<i>Bacteroides fragilis</i>	16	1	2	8	1	0.5
<i>Bacteroides fragilis</i> group	32	4	4	16	1	1
<i>Clostridium difficile</i>	16	2	8	8	2	8
<i>Clostridium perfringens</i>	1	0.5	0.12	1	0.5	0.25
<i>Fusobacterium</i> spp	8	1	2	4	0.5	4
<i>Peptostreptococcus</i> spp	4	1	0.12	2	0.5	1

MIC₉₀ = minimum inhibitory concentration required to inhibit 90% of isolates; Cipro = ciprofloxacin; Gati = gatifloxacin; Gemi = gemifloxacin; Levo = levofloxacin; Moxi = moxifloxacin; Trova = trovafloxacin. Adapted with permission (15).

KEY POINT

Bystander effects are important because of their association with the development of resistance through the selection of resistant clones and the replacement of the normal flora by potentially pathogenic organisms.

fluoroquinolones, a number of unanswered questions remain. For example, should fluoroquinolones with anaerobic activity, such as gatifloxacin and moxifloxacin, be used as single agents for mixed aerobic and anaerobic infections (eg, diabetic foot infections) or should they be given in combination with metronidazole? Disadvantages in using anaerobic fluoroquinolones as single agents include the possibility of bacterial resistance and the inhibition of the normal anaerobic flora, leading to overgrowth of other pathogens or colonization with potential pathogens.

Antibiotic use is associated with the development of vancomycin-resistant enterococci (VRE) (17). Evidence has shown that the use of antianaerobic agents may select for VRE. One recent study conducted >10 years in an academic medical center investigated the impact of restricting vancomycin and third-generation cephalosporins on prevalence of VRE (18). Rates of VRE increased despite restriction of vancomycin and third-generation cephalosporins, and clindamycin use significantly correlated with VRE, confirming that antianaerobic agents may predispose to VRE spread.

Similar ecologic issues arise with the new fluoroquinolones when treating respiratory or urinary tract infections. Are there potentially negative consequences to using anaerobic fluoroquinolones that may kill anaerobic colonic flora (*B fragilis*)?

To more fully understand this problem, we have constructed a colon model of a sick hospitalized patient to assess the influence of anaerobic treatment on *B fragilis*, *Klebsiella* species and other Enterobacteriaceae, *C difficile*, VRE, and *Candida albicans*. The model consists of serum vials with growth medium under anaerobic conditions and allows interactions of organisms and antimicrobials

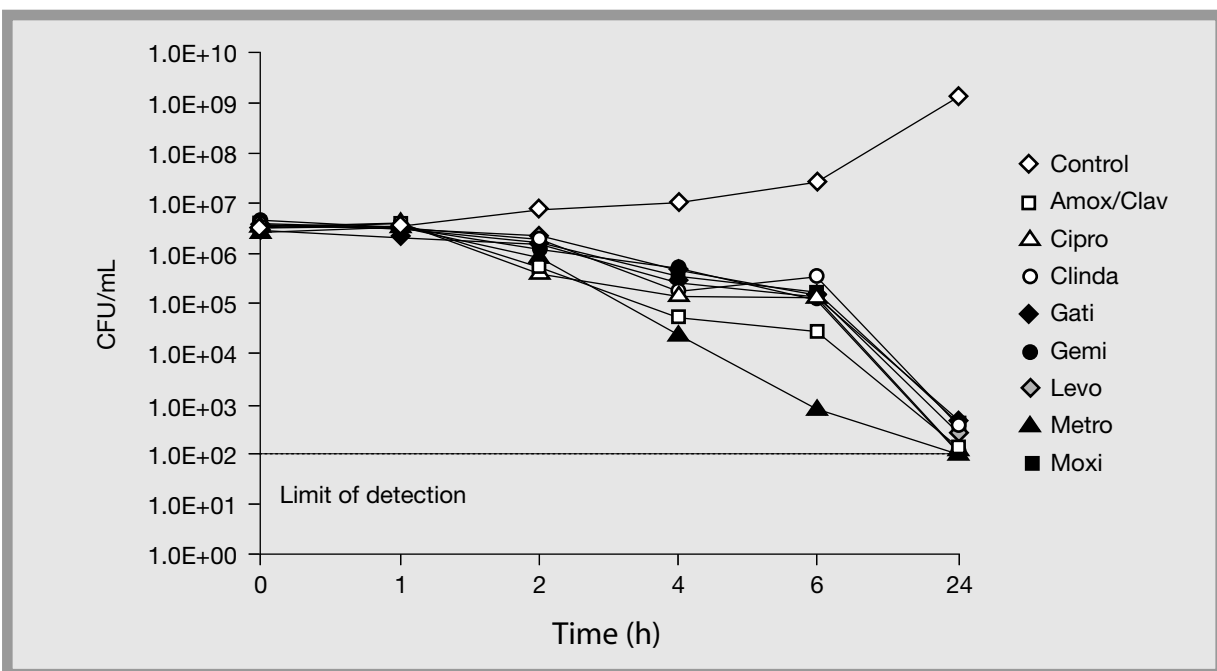


Figure 1. Kill curve of *Bacteroides fragilis* 11797 ($4 \times \text{MIC}$). (16) MIC = minimum inhibitory concentration; CFU = colony-forming unit; Amox/Clav = amoxicillin/clavulanate; Cipro = ciprofloxacin; Clinda = clindamycin; Gati = gatifloxacin; Gemi = gemifloxacin; Levo = levofloxacin; Metro = metronidazole; Moxi = moxifloxacin.

to be studied in a dynamic state. The model is continually refined. It is intended to mimic a sick, at-risk, hospitalized patient's bowel flora, which can be in a constant state of flux. The model enables us to vary parameters to understand the effect of exposure of organisms to various antimicrobials. Using this model to examine the effect of ciprofloxacin or levofloxacin (2 nonanaerobic agents) on *B fragilis*, *C albicans*, VRE, *K pneumoniae*, and *C difficile*, a slight fall in microbial levels is observed, which stabilizes around the limit of detection (**Figure 2**) (16). With gatifloxacin and moxifloxacin, which are more typically associated with activity against anaerobes, *B fragilis* falls within 24 hours; however, VRE significantly increases from 10^4 to 10^8 CFU/mL and *C albicans* slightly increases (**Figure 3**) (16). This model begins to explain how patients may be affected by various agents. The effects of these agents may be related to their concentration in the gut, which depends on how they are eliminated in the host, whether by hepatic or renal excretion. Antimicrobials in high-enough concentrations in the bowel can affect gut organisms, including those typically thought of as resistant based on MIC studies in the laboratory.

To illustrate this point, laboratory studies of maximum serum concentrations have demonstrated that serum levels were greatest for levofloxacin ($3.5 \mu\text{g/mL}$) compared with gatifloxacin ($2.8 \mu\text{g/mL}$), moxifloxacin ($1.5 \mu\text{g/mL}$), and ciprofloxacin ($1.4 \mu\text{g/mL}$). When we repeated these studies using colonic concentrations, a different pattern emerged with ciprofloxacin and moxifloxacin achieving much higher concentrations ($>1000 \mu\text{g/g}$ feces) compared with gatifloxacin or levofloxacin ($<100 \mu\text{g/g}$ feces) (15,19–22). Similar effects have been observed with *E coli* and macrolides such as clarithromycin. Despite laboratory results showing that it is resistant to clarithromycin, *E coli* is eradicated when clarithromycin antibiotic concentrations reach high levels of 300 to 400 $\mu\text{g/g}$ feces. In vitro activity does not always correlate with true effects in the bowel, and this should be considered when evaluating bystander effects.

These findings translate into effects in patients. In a study by Donskey et al (23), 51 patients who had been colonized with VRE were followed over a 7-month period to observe the effects of antibiotic treatment on the bowel (**Figure 4**). Patients who received an antianaerobic agent such as clindamycin or trovafloxacin had an increase in VRE density in the stool, whereas

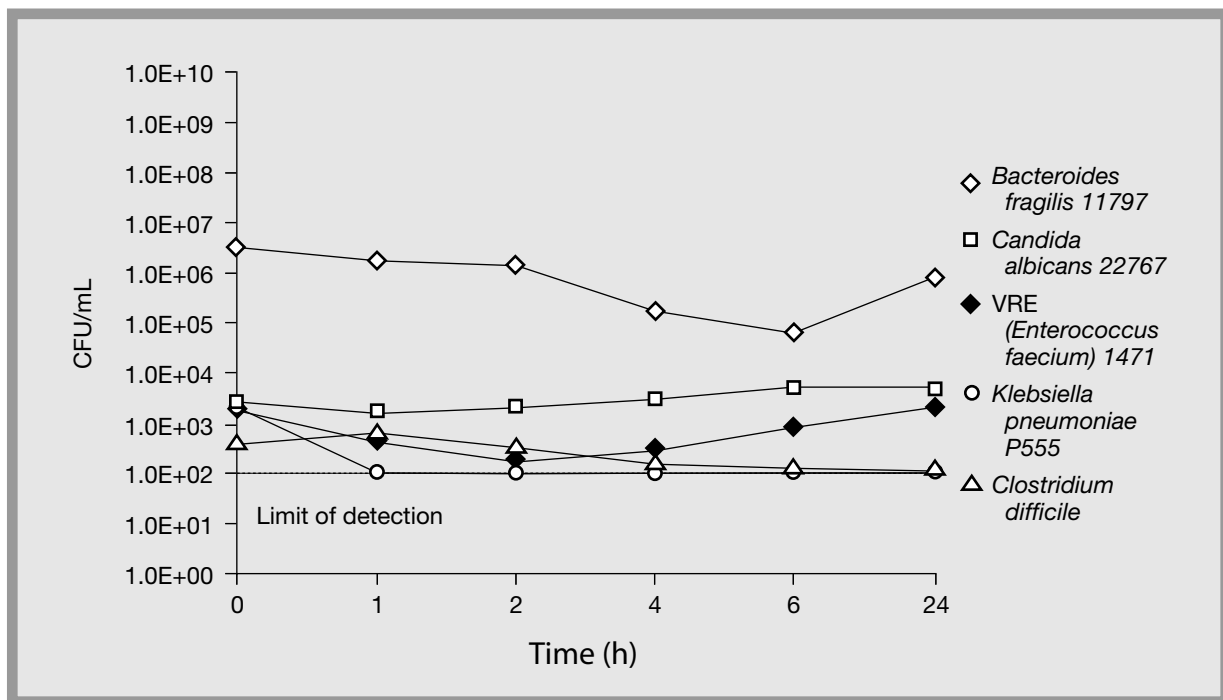


Figure 2. Exposure of colonic flora to nonanaerobic agents: ciprofloxacin 500 mg BID or levofloxacin 500 mg QD (16). CFU = colony-forming unit; VRE = vancomycin-resistant enterococci.

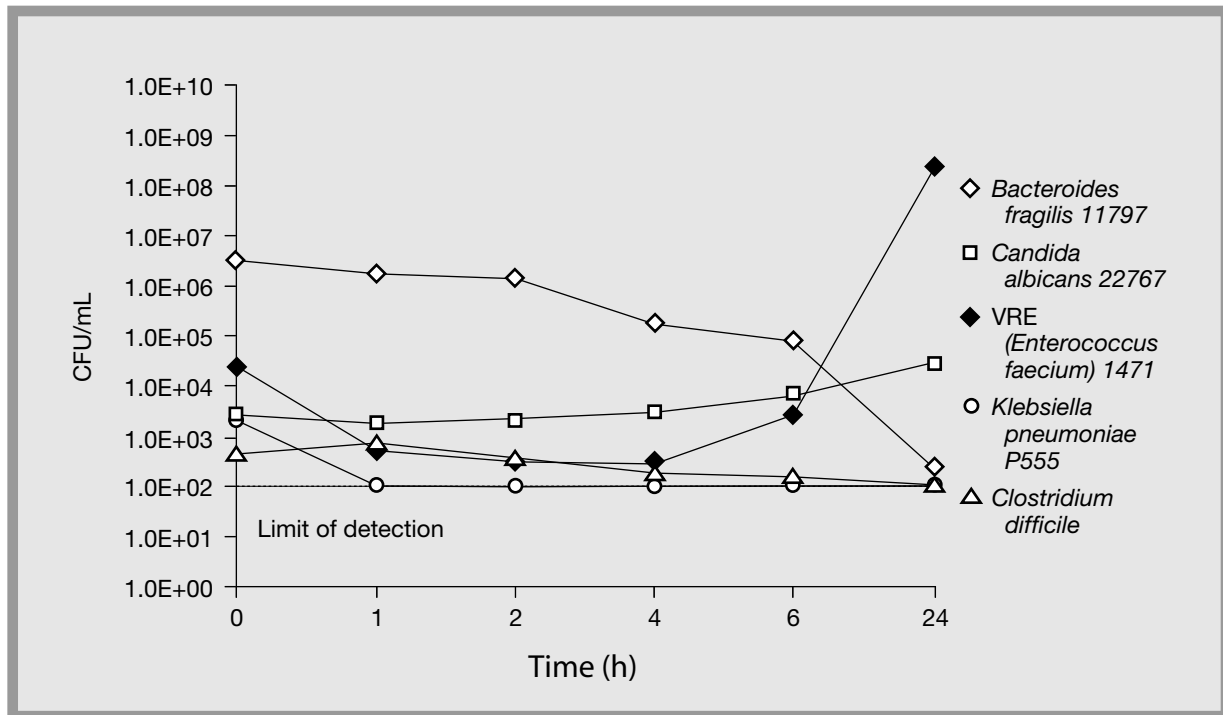


Figure 3. Exposure of colonic flora to anaerobic agents: gatifloxacin 400 mg QD or moxifloxacin 400 mg QD (16). CFU = colony-forming unit; VRE = vancomycin-resistant enterococci.

patients who were treated with an antianaerobic active agent with mild to minimal activity, such as ciprofloxacin or levofloxacin, had a drop in VRE concentrations in the stool. Thus, patients already colonized with VRE and who received an anaerobic antimicrobial had their concentrations of VRE increase, while patients who received a nonanaerobic antibiotic had their VRE concentrations drop. Patients with high VRE concentrations may spread their VRE more efficiently than those with lower VRE concentrations.

KEY POINT

Evidence demonstrating a correlation between antibiotic use and collateral damage to human intestinal and oral flora is lacking. However, ongoing studies are leading to a better understanding of how antimicrobials act and their collateral effects.

C difficile-associated diarrhea (CDAD) is another important complication of antibiotic use (24). It is normally associated with antianaerobic agents but is increasingly being observed with other antimicrobials. Ampicillin, the cephalosporins, clindamycin, and other broad-spectrum antibiotics are typically associated with CDAD in the literature. Fluoroquinolones have occasionally been reported to cause CDAD, whereas antianaerobic quinolones have not been clearly associated with *C difficile* colonization or infection. In one study conducted in the United Kingdom (25), the incidence of CDAD was observed for >5 years following a change in antimicrobial policy in 2 hospitals. The number of *C difficile*-positive specimens per 3-month period increased from 16 to 39, 9 months after switching from cefotaxime to ceftriaxone for treatment of pneumonia or septicemia. Following a change from ceftriaxone to levofloxacin, CDAD cases decreased from 1.46 to 0.34 cases per 1000 occupied bed days (Table IV) (25). The authors concluded that there appeared to be a strong relationship between treatment with ceftriaxone and

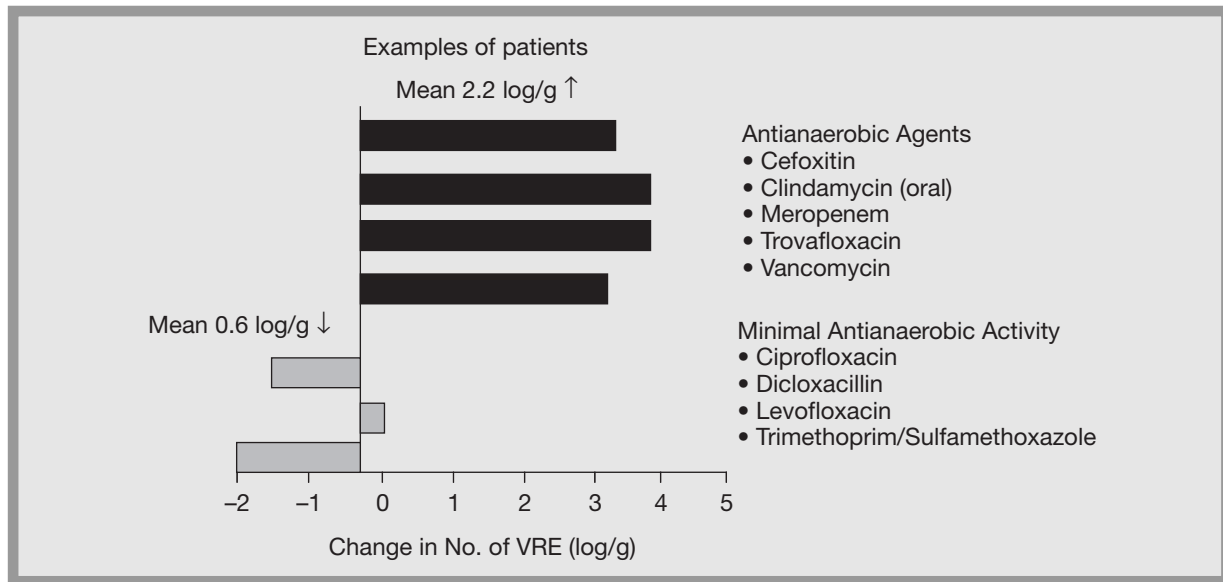


Figure 4. Effect of antibiotic therapy on the density of vancomycin-resistant enterococci (VRE) in stool of colonized patients. Adapted with permission (23).

CDAD incidence, and although this paper suggests an association, it is not possible to infer cause and effect from this study methodology.

Rao et al (26) compared the incidence of CDAD after treatment of LRTI with either levofloxacin or a beta-lactam (with or without a macrolide); 490 patients were treated with levofloxacin versus 448 patients with a beta-lactam (with a macrolide if necessary). Overall, the incidence of CDAD was 3.8%. The incidence was significantly greater with the beta-lactam (\pm macrolide) combination than with levofloxacin (5.6% vs 2.2%; $P < 0.01$). Length of hospital stay was also shorter in the levofloxacin arm ($P < 0.01$). The authors concluded that there were lower rates of

CDAD and a shorter length of inpatient stay by using levofloxacin for LRTIs.

A pattern was presented at a recent conference (27). A tertiary care hospital reported a high rate of nosocomial CDAD with an incidence of 1.5 per 1000 patient days. Infection control measures and restricted use of clindamycin failed to impact the CDAD rate. A change in the antibiotic treatment pattern, with decreased use of cephalosporins and increased use of ticarcillin/clavulanate and fluoroquinolone, resulted in an unexpected reduction in the CDAD rate to 0.6 per 1000 patient days.

CONCLUSIONS

The use of antianaerobic antibiotics is associated

TABLE IV. ANTIMICROBIAL USE AND CDAD

Purpose: Analyze incidence of CDAD with changing antibiotic policy at 2 hospitals over 5 years

CDAD cases jump 9 months after switch from cefotaxime to ceftriaxone, from 16 to 39 *C difficile*-positive samples per quarter

After switch from ceftriaxone to levofloxacin, cases of CDAD decreased, from 1.46 to 0.34 cases per 1000 occupied bed days

Conclusion: Strong relationship between ceftriaxone treatment and CDAD incidence

CDAD = *Clostridium difficile*-associated diarrhea. Adapted with permission (25).

with the unintended consequence of collateral damage to the normal bowel flora. Some of these effects are theoretical at this stage of knowledge. As we gradually develop a theory, we are beginning to ask relevant questions, develop and test hypotheses, formulate policies, and better understand not just how and where antimicrobials act, but how bystander effects may become clinically important. These effects are important because of their association not only with the development of resistance through the selection of resistant clones but also with the replacement of the normal flora by potentially pathogenic organisms. Prolonged VRE colonization, increased *C difficile* burden of infection and, more recently, increased rates of *C albicans* proliferation are all possible consequences of collateral damage. These organisms and others are worthy of our study as we determine the collateral effects of antimicrobials.

ACKNOWLEDGMENT

Assistance in the writing of this article was provided by Dr. A. Niroshan Siriwardena, MB, MMedSci, PhD, FRCGP, Family Physician and Clinical Senior Lecturer, De Montfort University, Leicester, UK.

Address correspondence to: Daryl J. Hoban, PhD, Health Sciences Centre, 820 Sherbrook Street, Clin Micro – MS673 #6640, Winnipeg, Manitoba, Canada R3A 1R9. Phone: (204) 787-1191. Fax: (204) 787-4699. E-mail: dhoban@hsc.mb.ca

REFERENCES

1. Marshall JC. Gastrointestinal flora and its alterations in critical illness. *Curr Opin Clin Nutr Metab Care*. 1999;2:405–411.
2. Gould IM. Antibiotic policies and control of resistance. *Curr Opin Infect Dis*. 2002;15:395–400.
3. Kariuki S, Hart CA. Global aspects of antimicrobial-resistant enteric bacteria. *Curr Opin Infect Dis*. 2001;14:579–586.
4. Andremont A. The future control of bacterial resistance to antimicrobial agents. *Am J Infect Control*. 2001;29:256–258.
5. Andersson MI, MacGowan AP. Development of the quinolones. *J Antimicrob Chemother*. 2003; 51(Suppl 1):1–11.
6. Emmerson AM, Jones AM. The quinolones: decades of development and use. *J Antimicrob Chemother*. 2003;51(Suppl 1):13–20.
7. Bartlett JG, Dowell SF, File TMJ, et al. Practice guidelines for the management of community-acquired pneumonia in adults. *Clin Infect Dis*. 2000;31:347–382.
8. Lautenbach E, Larosa LA, Kasbekar N, et al. Fluoroquinolone utilization in the emergency departments of academic medical centers: prevalence of, and risk factors for, inappropriate use. *Arch Intern Med*. 2003;163:601–605.
9. Martin SJ, Jung R, Garvin CG. A risk-benefit assessment of levofloxacin in respiratory, skin and skin structure, and urinary tract infections. *Drug Saf*. 2001;24:199–222.
10. Lautenbach E, Fishman NO, Bilker WB, et al. Risk factors for fluoroquinolone resistance in nosocomial *Escherichia coli* and *Klebsiella pneumoniae* infections. *Arch Intern Med*. 2002;162:2469–2477.
11. Davies TA, Goldschmidt R, Pflieger S, et al. Cross-resistance, relatedness and allele analysis of fluoroquinolone-resistant US clinical isolates of *Streptococcus pneumoniae* (1998–2000). *J Antimicrob Chemother*. 2003;52:168–175.
12. Niederman MS, Mandell LA, Anzueto A, et al. Guidelines for the management of adults with community-acquired pneumonia. *Am J Respir Crit Care Med*. 2001;163:1730–1754.
13. British Thoracic Society Standards of Care Committee. BTS guidelines for the management of community acquired pneumonia in adults. *Thorax*. 2001;56(Suppl 4):IV1–64.
14. Scheld WM. Maintaining fluoroquinolone class efficacy: review of influencing factors. *Emerg Infect Dis*. 2003;9:1–9.
15. Zhanel GG, Ennis K, Vercaigne L, et al. A critical review of the fluoroquinolones: focus on respiratory infections. *Drugs*. 2002;62:13–59.
16. Zhanel GG, et al. Presented at: The 102nd General Meeting of the American Society for Microbiology; May 19–23, 2002; Salt Lake City, Utah.
17. Dever LL, China C, Eng RH, et al. Vancomycin-resistant *Enterococcus faecium* in a Veterans Affairs medical center: association with antibiotic usage. *Am J Infect Control*. 1998;26:40–46.
18. Lautenbach E, LaRosa LA, Marr AM, et al. Changes in the prevalence of vancomycin-resistant enterococci in response to antimicrobial formulary interventions: impact of progressive restrictions on use of vancomycin and third-generation cephalosporins. *Clin Infect Dis*. 2003;36:440–446.
19. Avelox [package insert]. West Haven, Conn: Bayer Corporation; 2003.
20. Cipro [package insert]. West Haven, Conn: Bayer Corporation; 2003.
21. Levaquin [package insert]. Raritan, NJ: Ortho-McNeil Pharmaceutical, Inc; 2003.
22. Wilcox MH, Fawley W, Freeman J, et al. In vitro activity of new generation fluoroquinolones against genotypically distinct and indistinguishable *Clostridium difficile* isolates. *J Antimicrob Chemother*. 2000;46:551–556.

23. Donskey CJ, Chowdhry TK, Hecker MT, et al. Effect of antibiotic therapy on the density of vancomycin-resistant enterococci in the stool of colonized patients. *N Engl J Med.* 2000;343:1925–1932.
24. McFarland LV. Epidemiology of infectious and iatrogenic nosocomial diarrhea in a cohort of general medicine patients. *Am J Infect Control.* 1995; 23:295–305.
25. 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;54:104–108.
26. Rao Gopal G, Rao Mahankali CS, Starke I. *Clostridium difficile*-associated diarrhea in patients with community-acquired lower respiratory infection being treated with levofloxacin compared with β -lactam-based therapy. *J Antimicrob Chemother.* 2003;51:697–701.
27. Miller M, et al. *Clostridium difficile*-associated diarrhea and antimicrobial use. Presented at: The 42nd Interscience Conference on Antimicrobial Agents and Chemotherapy; September 27–30, 2002; San Diego, Calif.

Clinical Cornerstone® Monograph. Copyright © 2004 Excerpta Medica, Inc.