**Clostridium difficile** colitis that fails conventional metronidazole therapy: response to nitazoxanide

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**Objectives:** Clostridium difficile-associated disease has increased in incidence and severity. Recommended treatments include metronidazole and vancomycin. Recent investigations, however, document the failure of metronidazole to cure a substantial proportion of patients with Clostridium difficile colitis, but oral administration of vancomycin raises concerns over selection of antibiotic-resistant organisms in the hospital environment. We have recently shown that nitazoxanide is as effective as metronidazole in initial therapy for C. difficile colitis. We hypothesized that this drug might be effective in treating patients who fail therapy with metronidazole.

**Methods:** In the present study, we identified 35 patients who failed treatment with metronidazole for C. difficile colitis; failure was defined as either no improvement in symptoms or signs of disease (28 patients) after ≥14 days of treatment with metronidazole or prompt recurrence on at least two occasions after initially responding to such treatment (seven patients). These patients were ill with numerous co-morbidities. Nitazoxanide, 500 mg twice daily, was given for 10 days; results from all patients are included.

**Results:** Twenty-six (74%) of 35 patients responded to nitazoxanide, of whom seven later had recurrent disease, yielding a cure rate of 19 of 35 (54%) from initial therapy. Three who initially failed and one who had recurrent disease were re-treated with, and responded to, nitazoxanide. Thus, the aggregate cure with nitazoxanide in this difficult-to-treat population was 23 of 35 (66%).

**Conclusions:** Nitazoxanide appears to provide effective therapy for patients with C. difficile colitis who fail treatment with metronidazole.

Keywords: antibiotic-associated diarrhoea, vancomycin, C. difficile

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**Introduction**

Clostridium difficile-associated disease has emerged as a major nosocomial infection in North American and European hospitals, with a rising incidence, increasing severity and an increasing rate of failure to conventional therapy.1–4 Vancomycin was the first drug shown to provide effective treatment for this condition5 and, at least in the USA, remains the only one approved for this purpose by the Food and Drug Administration.6 Early studies, however, appeared to show metronidazole to be equally effective.6–8 Initially driven by the prohibitive cost of oral vancomycin, and subsequently by fear of selecting vancomycin-resistant bacteria in hospitals, health-care providers have tended to prescribe metronidazole, which is now recommended as first-line therapy for this condition,9,10 even though the response to vancomycin is somewhat more rapid.11 We4 and others3,12 have recently documented a substantial rate of failure with this therapy. When treatment with metronidazole fails, most infectious disease specialists13,14 recommend oral vancomycin, but continuing concerns over the high cost and the emergence of vancomycin-resistant bacteria, reinforced by federal guidelines discouraging excessive use of this medication,15 have motivated the search for alternative therapy.
Nitazoxanide, a nitrothiazolide, successfully treats infestation by intestinal parasites and is approved in the USA and UK to treat cryptosporidiosis and giardiasis. This drug acts by blocking anaerobic metabolism. In vitro, low concentrations of nitazoxanide or its breakdown product, tizoxanide, inhibit C. difficile. Two-thirds of an administered dose is excreted in the faeces. In a prospective, double-blind study, we recently found that nitazoxanide is at least as effective as metronidazole in treating patients with moderately severe or severe C. difficile colitis. Taken together, these factors motivated us to study nitazoxanide in patients with C. difficile colitis who had failed conventional therapy with metronidazole. Interestingly, Bartlett also suggested this possibility in a recent editorial on the subject.

**Patients and methods**

**Study site**

This study was carried out at the Michael E. DeBakey Veterans Affairs Medical Center, Houston. Patients in this system generally rely exclusively on this center for their medical care. Medical and laboratory records are fully automated, and every encounter between a patient and the medical system is recorded electronically. Thus, follow-up data tend to be remarkably complete. During the time of this study, the therapy approved in our hospital for treating C. difficile colitis was metronidazole. For reasons summarized above, vancomycin was used infrequently, and an order to do so required approval by a member of the Infectious Disease Section.

**Patients**

Patients who had clearly failed conventional treatment for C. difficile colitis were asked to participate in an IRB-approved open-label study of nitazoxanide (Institutional Review Board, Baylor College of Medicine, Protocol H-15601). Therapeutic failure was defined as either: (i) the absence of a response to metronidazole and/or oral vancomycin therapy, defined as persistence of fever, diarrhoea (at least three loose or watery stools per day), abdominal pain and/or otherwise unexplained leucocytosis and a positive EIA for C. difficile toxin after 14 days of oral treatment with a total dosage of 1.5 g of oral metronidazole or 500 mg of oral vancomycin daily; or (ii) on at least two occasions, an initial response to metronidazole or vancomycin, as defined by resolution of the above-named symptoms, followed within 30 days by recurrence. Because metronidazole may be effective in treating an initial recurrence of disease, in order to be included in this study, patients needed to have at least two bouts of recurrent symptoms and signs of colitis after a full course of treatment. Symptoms were present for ≥4 days before the diagnosis of recurrent disease was made. Nitazoxanide, 500 mg, was given twice daily, for 10 days. Initially, informed consent was sought in accordance with IRB guidelines. After the first 22 patients were treated, the results of the double-blind study comparing metronidazole to nitazoxanide were revealed, and nitazoxanide was placed on the hospital formulary, requiring approval for its use by one of the investigators (DMM or RJH). Thereafter, 13 additional patients who failed conventional therapy were treated with nitazoxanide but without informed consent being required. All patients were followed closely throughout their treatment and thereafter for 60 days in order to determine whether there was a recurrence. A separate IRB-approved protocol (Baylor IRB H-16175) authorized a review of all medical records after treatment had been completed. Data presented in this paper are from 35 consecutive patients who met the above criteria and were treated with nitazoxanide; results from all patients are included.

**Response to treatment**

Patients were seen regularly by an investigator after treatment with nitazoxanide was begun. Interviews with patients, family members and nurses were used to determine the number and quality of stools, and the persistence of fever, abdominal discomfort and/or leucocytosis was noted. Failure of therapy was defined by the persistence of at least three loose or watery stools per 24 h, along with fever, leucocytosis, abdominal pain and/or urgency. A response to treatment was defined as the absence of symptoms of disease as defined above. After the study was over, complete electronic medical records were reviewed for ≥60 days to verify all recorded information and to determine whether there had been a recurrence or possible recurrence of C. difficile colitis. Patients were said to have undergone an apparent cure when, after a response, there was no recurrence of symptoms or signs of colitis during the ensuing 60 days. An initial response to nitazoxanide followed by a return of these symptoms and signs together with a positive faecal C. difficile toxin assay was termed a recurrence.

**Microbiology**

C. difficile toxin was detected by EIA (Premier™ Toxins A&B, Meridian Bioscience, Cincinnati, OH, USA). Faecal samples from some patients were submitted in anaerobic transport containers to Microbiology Specialists Incorporated (Houston, TX, USA) for anaerobic culture. Definitive identification of C. difficile was by distinctive isoacid production on gas liquid chromatography. Susceptibilities to vancomycin, metronidazole, nitazoxanide and tizoxanide were studied by agar dilution. Isolates were sent to the Centers for Disease Control and Prevention (Atlanta, GA, USA) where Dr Charles E. Kilgore, Ms Angela Thompson and Dr L. Clifford McDonald graciously performed PFGE, toxinotyping and detection of binary toxin production.

**Results**

**Demographics**

Thirty-five consecutive patients who were treated with nitazoxanide after failing conventional therapy for C. difficile colitis are included in this report. Thirty-three were male and two were female, consistent with gender distribution at our hospital. The mean age was 67.5 years (range 49–86). In 28 patients, symptoms and signs of C. difficile colitis had persisted without response despite treatment with metronidazole for 14 days; seven had at least two recurrences after appropriate treatment (Figure 1). Those with persistent disease had been treated with metronidazole for 17–45 days (mean = 22.4 days, median = 21 days); two also
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There appeared to be no differences in response to nitazoxanide among patients who had failed metronidazole and those who had recurrence of disease after initial responses. Thus, for ease of presentation, results are merged for the two groups, although Figure 2 shows actual outcomes for all 35 patients. Twenty-six of the 35 (74%) patients had a rapid resolution of symptoms and signs of colitis after nitazoxanide therapy was begun (Figure 2), including 20 of the 28 (71%) who had persisting disease despite metronidazole and 6 of the 7 (86%) who had recurrent disease (difference not significant, \( P = 0.65 \), Fisher’s exact test). Diarrhoea stopped within a mean of 2.6 days (range 1–4). Febrile patients defervesced and leucocytosis, when attributable to the colitis, resolved within 4 days. Sixty days after treatment, 19 of these 26 (73%; 54% of the original 35 patients) remained free of symptoms or signs of colitis and were regarded as apparent cures.

Seven of the 26 patients who initially responded to nitazoxanide (27%; 20% of the original 35 patients) had recurrent *C. difficile* colitis within a mean of 12.1 (range 7–24) days after the completion of therapy (Figure 2). After the initial course of therapy with nitazoxanide, treatment of any subsequent failure or recurrence was selected by the primary care providers. Of these seven patients with recurrent disease, one was re-treated with nitazoxanide and responded. Four were treated with metronidazole, of whom two were cured and two failed to respond (one of these received vancomycin and metronidazole together without response). Two were treated with vancomycin of whom one initially responded but died before completion of therapy and one failed, eventually dying with persistent *C. difficile* colitis.

Nine of the 35 (26%) patients failed nitazoxanide (Figure 2). One died in the first week of therapy. Two were re-treated with metronidazole but failed to respond. One each of these was subsequently treated successfully with an additional course of nitazoxanide or vancomycin (responding only to a second course of this drug). Three were re-treated with nitazoxanide; one was cured, one was eventually cured after two additional courses of nitazoxanide, and one did not respond. Three were treated with long courses of vancomycin, but had persisting symptoms and signs of colitis until they died. Thus, in total, 23 of 35 (66%) patients with *C. difficile* colitis who had failed metronidazole were eventually cured by nitazoxanide.

**Microbiology**

*C. difficile* was isolated from faecal samples of 15 patients. By agar dilution, all isolates were inhibited by metronidazole (median MIC = 1 mg/L, range 0.25–2 mg/L), vancomycin (median MIC 4 mg/L, range 1–8 mg/L), nitazoxanide (median MIC 1, range 0.25–2 mg/L) and tizoxanide (median MIC 0.5 mg/L, range 0.25–1 mg/L). In order to determine whether persistent or recurrent infection was due to a new, virulent strain of *C. difficile* we submitted 13 retrievable isolates to the Epidemiology and Laboratory Branch of the Centers for Disease Control and Prevention (Dr G. E. Kilgore, Ms A. Thompson and Dr L. C. McDonald). Two of the 13 (15%) were identical with the recently identified epidemic, toxin gene-variant strain, PFGE type NAP-1, toxinotype III, binary toxin positive (see Figure 3). Eight (62%) isolates were PFGE type NAP-2,
toxinotype 0 and binary toxin negative. The remaining three (23%) isolates were of as-yet unnamed PFGE types.

Discussion

Nosocomial *C. difficile* colitis has evolved into a major problem in developed countries. Recent studies indicate that as many as 20–25% of patients with this condition fail therapy with metronidazole, and another 20–25% have recurrent disease, substantially higher rates of failure than were previously reported. The usual therapy for patients who fail metronidazole has been oral vancomycin, but there is great reluctance to prescribe this drug because of concern for selection of vancomycin-resistant bacteria. Nitazoxanide has recently been shown to be at least as effective as metronidazole in the initial treatment of moderately severe to severe *C. difficile* colitis. Taken together, these factors motivated us to study nitazoxanide in treating patients who fail conventional therapy for this infection.

We identified 35 patients in whom *C. difficile* colitis had plainly failed treatment with metronidazole; 28 of these had persistent symptoms of colitis despite prolonged therapy with

![Flow sheet showing all patients studied and the status of their *C. difficile* infection at each stage in the study. Nitazox, nitazoxanide; metronid, metronidazole; vanco, vancomycin.](image)

![PFGE of *C. difficile* isolated from 13 patients in this study. The figure shows that no single strain predominated. Eight of 13 isolates were NAP-2, toxinotype 0, and binary toxin negative, representing several different subtypes within PFGE NAP-2. Only two isolates were identified as the newly emerging NAP-1, toxinotype III, binary toxin positive epidemic strain.](image)
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metronidazole, and seven had at least two relapses within a few weeks of an apparent response. A prompt response to nitazoxanide was seen in 26 (74%) cases, including 20 of the 28 (71%) who had persisting colitis despite metronidazole and six of the seven (86%) who had recurrent disease.

Of the 26 who responded to nitazoxanide, seven had recurrent *Clostridium difficile* colitis, one of whom eventually responded to nitazoxanide. Of nine whose symptoms persisted during initial treatment with nitazoxanide, three were eventually cured by repeated course(s) of this drug. Thus, in aggregate, nitazoxanide produced a cure in 23 of 35 (66%) patients who had already failed other therapy, a response rate similar to that observed in initial treatment of unselected *Clostridium difficile* colitis hospitalized patients with metronidazole. Achieving this rate of response in ill patients who had already failed prior therapy indicates a potentially important role for nitazoxanide in treating this disease.

PFGE showed that the majority of the strains studied were types other than the recently described NAP-1, toxinoftype III, binary toxin positive isolate epidemic strain. This finding indicates that the new, epidemic strain is not solely responsible for the recent increase in incidence and severity of *Clostridium difficile* colitis in hospitalized patients in the USA. In our patients, the high failure rate appears to be more closely related to host factors, reflecting a population that is older and sicker than in the past, and that has received more broad coverage antibiotic therapy. More widespread usage of proton pump inhibitors may also play a contributory role. Co-morbidities were prominent in our patients, and seven (20%) died within the 60 day study period. Further support for the central role of the host response in recurrent disease comes from our preliminary observation that most of our patients had low levels of antibody to toxins A and B both acutely and in follow-up, a finding that is consistent with susceptibility to recurrent disease. Antimicrobial resistance of *Clostridium difficile* did not contribute to failure, because isolates from our patients were all susceptible to metronidazole, vancomycin, nitazoxanide and its metabolic product tizoxanide.

The chief limitation of this study is that, although prospective, it was open and non-comparative. At the time it was undertaken, there were precious few possible treatments, as emphasized by Louie in his recent editorial. It is always possible that these patients might readily have responded to an additional course of conventional therapy. D. Gerding has shown (unpublished, presented at Symposium on *Clostridium difficile*, Annual Meeting Infectious Disease Society of America, 12–15 October 2006) that some patients respond to metronidazole more slowly than was previously thought, and Pepin et al. have reported that metronidazole may bring about a response in one-third of patients who are re-treated for a recurrence following therapy with that drug. That is why our inclusion criteria were so stringent. Patients who failed metronidazole had been treated for at least 14 days (mean duration of treatment was 22.4 days), and those with recurrent disease all had at least two recurrences (mean number of recurrences, 2.6).

We have reviewed elsewhere the use of other therapeutic modalities for *Clostridium difficile* colitis, including biological agents such as non-toxigenic *C. difficile* or other non-toxigenic bowel flora, antimicrobial agents such as bacitracin, teicoplanin and rifaximin, toxin-binding polymers, immune globulin or monoclonal antibody to *C. difficile* toxin, and non-steroidal anti-inflammatory drugs. As always, prevention is better than treatment, and continued emphasis on judicious use of antibiotics and appropriate infection control measures remain central to the art of good medical practice.

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Transparency declarations

This study was conceived and designed by the investigators who initiated the proposal and received funding to the Infectious Disease Section from Romark Pharmaceuticals. This company is privately owned, and none of the participants in this study has any share in the company or its profits. D. M. M. has received travel expenses and an honorarium for his appearance at a satellite symposium.

References