Nitazoxanide versus Vancomycin in *Clostridium difficile* Infection: A Randomized, Double-Blind Study

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Background. Vancomycin is the only US Food and Drug Administration–approved drug for treatment of *Clostridium difficile* infection (CDI). Metronidazole has been widely used for this purpose but may be inferior to vancomycin, especially for hospitalized patients with severe disease. We report a prospective, double-blind, randomized controlled trial comparing nitazoxanide with vancomycin for treatment of CDI.

Methods. Fifty patients with CDI were randomized to receive vancomycin or nitazoxanide for 10 days. An initial response was considered to be the absence of all CDI symptoms between days 11 and 13, and a final response was considered to be lack of symptom recurrence by day 31.

Results. One patient fulfilled an exclusion criterion and was removed from the study. Twenty-seven patients received vancomycin, and 23 received nitazoxanide; 23 and 18 patients, respectively, completed the full course of treatment. Initial responses occurred in 20 (74%) of 27 patients treated with vancomycin and in 17 (77%) of 22 patients treated with nitazoxanide (95% confidence interval, −24% to +28%). In those who completed therapy, response rates were 87% (20 of 23 patients) in the vancomycin group and 94% (17 of 18 patients) in the nitazoxanide group (95% confidence interval, −18% to +30%). Times to complete resolution of symptoms were similar in the 2 groups ($P = .55$). Two patients in the vancomycin group and 1 patient in the nitazoxanide group experienced relapse within 31 days after beginning treatment. Sustained response rates were 78% (18 of 23 patients) for the vancomycin group, and 89% (16 of 18 patients) for the nitazoxanide group (95% confidence interval, −18% to +35%).

Conclusions. The small sample precludes conclusions about noninferiority of nitazoxanide to vancomycin. Nevertheless, this is the first recent randomized controlled trial to compare any antimicrobial agent other than metronidazole with vancomycin. Results suggest that nitazoxanide may be as effective as vancomycin in treating CDI.

Trial registration. ClinicalTrials.gov identifier: NCT00384527.

The epidemic of *Clostridium difficile* infection (CDI) has expanded, and the disease has become more severe [1, 2]; according to a recent report, the incidence of CDI and the case fatality rate in the United States both doubled between 2000 and 2005 [3]. Vancomycin is the only drug that has been approved by the US Food and Drug Administration for treating CDI [4]. Since the mid-1980s, metronidazole has been widely used in preference to vancomycin, on the basis of studies that suggested equivalency of effect and because of concerns over excessive cost and selection of vancomycin-resistant bacteria [4, 5]. More-recent case series, however, have shown substantial failure rates associated with this drug [6, 7]. Two direct comparisons have shown metronidazole to be inferior to vancomycin in treating CDI, except in patients with mild disease [8, 9], although, somewhat paradoxically, a recent retrospective analysis has suggested that disease specifically due to the so-called epidemic or hypervirulent strain (BI/NAP1/027) may not respond better to vancomycin than to metronidazole [10].

Although a number of newer antimicrobial agents

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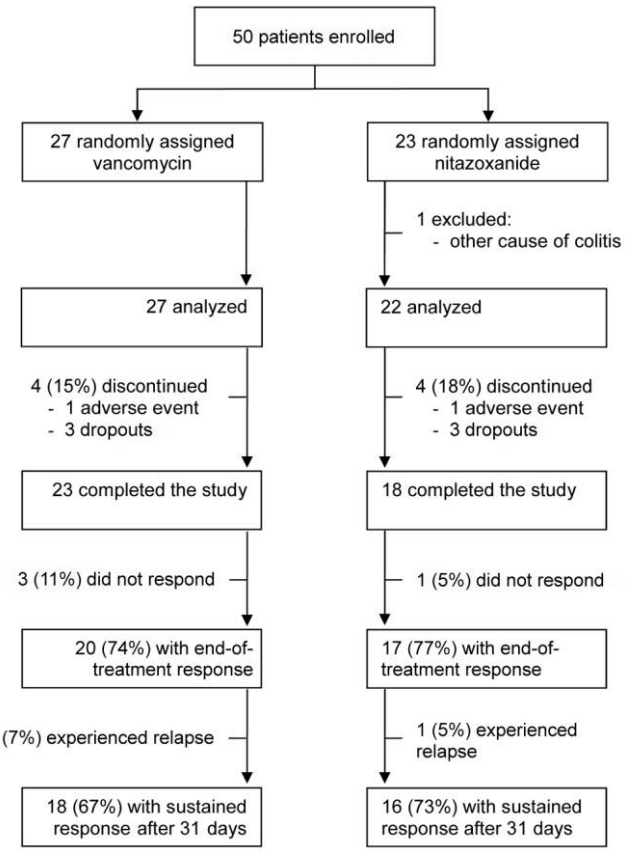

Figure 1. Flow chart showing enrollment of patients, numbers of patients who completed the protocol, and patient outcomes.

have been proposed [4, 5, 11], they have not been compared with vancomycin in prospective, double-blind, randomized controlled trials. We reported that nitazoxanide, a drug that interferes with anaerobic metabolism of protozoa and bacteria [12], is at least as effective as metronidazole in treating CDI [13] and also is effective for patients who experience treatment failure with metronidazole therapy [14]. We now report results from a prospective, double-blind, randomized controlled trial that compared vancomycin with nitazoxanide in patients with CDI.

PATIENTS AND METHODS

Patients. Patients were eligible for inclusion if they had a fecal sample submitted to the microbiology laboratory of a participating hospital that had EIA results positive for *C. difficile* toxin (Premier Toxins A & B; Meridian Bioscience). To be included in this study, in addition to test results positive for *C. difficile* toxin, patients needed to have ⩾3 loose stools within 24 h and ⩾1 of the following additional findings: fever (temperature, >38.3°C), abdominal pain, and/or leukocytosis. Patients were excluded if they (1) were unwilling or unable to participate, (2) had another condition that might cause diarrhea (such as inflammatory bowel disease), (3) were undergoing enteral tube feeding, (4) had >1 recurrence of CDI during the 6 months before being considered for enrollment, or (5) had used any drug with anti–*C. difficile* activity—with the exception of ⩽3 oral doses of metronidazole or vancomycin—within 1 week before being considered for enrollment in the study. We also excluded patients with unstable vital signs who were hospitalized in an intensive care unit, but we were careful not to exclude patients who were otherwise quite ill, to avoid selection bias toward patients with mild (and, therefore, potentially self-limited) disease. Cultures and microscopic examinations of fecal samples for ova and parasite were performed at baseline to exclude other infectious causes of diarrhea. Severe CDI was defined using a modification of the severity score recently described by Zar et al. [9]. One point each was assigned for age ⩾60 years, >7 stools/day, temperature >38.3° C, albumin level <2.5 gm/dL, or WBC count >15,000 cells/mm^3; a score of ⩾2 points was regarded as severe disease.

Assignment to regimen. Patients were asked to sign a consent form that had been approved by the local institutional review board at each of the 10 participating sites. Those who signed were randomized to receive either nitazoxanide (500 mg every 12 h) or vancomycin (125 mg every 6 h) for a total of 10 days. "Dummy" placebo tablets or capsules were dispensed, to assure that nursing and medical staff and patients were blind to patient study status. Packages of double-blind study medications and placebos were prepared by the study sponsor, Romark Laboratories, and were allocated to study sites in randomized blocks of 4. At the time of randomization, each site sequentially assigned each patient a number from its allotment of blinded study medication. The randomization code was sealed and maintained in the files of the study sponsor until all data on all patients had been submitted by individual investigators, and the database was locked in accordance with US Food and Drug Administration regulations. This study is registered at ClinicalTrials.gov (NCT00384527).

Definitions of response, failure, and relapse. After therapy was begun, patients were seen daily by one of the investigators for 31 days or until the time of hospital discharge. Attention was given specifically to the time from the beginning of treatment until the last loose stool, defervescence, and resolution of abdominal pain and leukocytosis. Patients who were discharged from the hospital maintained diaries documenting compliance with the treatment regimen, symptoms, and adverse events. They were seen between days 12 and 14 and were seen or contacted by telephone on day 31. The purposes of follow-up were to record the response to therapy at the 11–13 day end point, as well as to monitor adverse events and compliance with medication regimen. An end-of-treatment response was defined as complete resolution of all symptoms and signs attributable to CDI during the 3 days after completion of therapy.

Table 1. Demographic characteristics and comorbidities of study patients.

Variable	Vancomycin group (n = 27)	Nitazoxanide group (n = 22)	P
Ethnicity			
White	22 (81)	12 (55)	
Black	5 (19)	10 (45)	.06
Sex			
Male	18 (67)	14 (64)	
Female	9 (33)	8 (36)	.99
Age, mean years	65.7	59.6	.19
Inpatient	21 (78)	21 (95)	.11
Bedridden status	4 (15)	5 (23)	.71
Malnourished status (serum albumin ⩽3.4 g/dL)	18/22 (82)	17/19 (89)	.49
Neurological disease	8 (30)	7 (32)	.99
Diabetes mellitus	15 (56)	10 (45)	.57
Renal insufficiency	9 (33)	6 (27)	.76
Liver disease	3 (11)	5 (23)	.44
Severe *Clostridium difficile* infection	10 (37)	10 (45)	.57

NOTE. Data are no. (%) of patients, unless otherwise indicated.

A return of symptoms after an initial response but within 31 days after the onset of treatment was regarded as a relapse if *C. difficile* toxin was detected in stool samples by EIA or if the patient was re-treated empirically for CDI and responded to treatment. For the modified intent-to-treat analysis, the protocol listed as treatment failure any case that involved a patient who received another medication for CDI or who died before the completion of treatment.

Statistics. The protocol called for efficacy analysis of a modified intent-to-treat population that consisted of all enrolled patients randomized (excluding any patient who, after enrollment, was found to have fulfilled an exclusion criterion). The primary end point was the clinical response at the end of treatment. Secondary end points included time to resolution of symptoms and sustained response rate at day 31 (end-of-treatment response with no recurrence). The rates of response to vancomycin or nitazoxanide were calculated and compared by determining the 95% CI for the difference [15]. The times from the first dose of medication to the resolution of all symptoms of colitis were compared for each treatment group with use of the Kaplan-Meier survival analysis and a Prentice-modified Wilcoxon test, with $\alpha = .05$. The frequencies of adverse events by treatment group were compared using Fisher's exact test or the χ^2 test, as appropriate.

The study was designed to demonstrate noninferiority of nitazoxanide, compared with vancomycin, through use of a

Table 2. Clinical manifestations of *Clostridium difficile* disease at baseline.

Variable	Vancomycin group (n = 27)	Nitazoxanide group (n = 22)	P
Stool consistency			.42
Liquid	21 (78)	19 (86)	
Liquid or soft	4 (15)	3 (14)	
Soft	2 (7)	…	
Stool frequency, within 24 h			.59
3–4	11 (41)	10 (45)	
5–10	14 (52)	9 (41)	
>10	2 (7)	3 (14)	
Mean no. of stools in 24 h before treatment	5.9	6.5	.59
Fever	4 (15)	3 (14)	.99
Abdominal pain	21 (78)	19 (86)	.49
Peripheral leukocytosis	14 (52)	11 (50)	.99
C. difficile infection within 90 days before study enrollment	5 (19)	2 (9)	.43

NOTE. Data are no. (%) of patients, unless otherwise indicated.

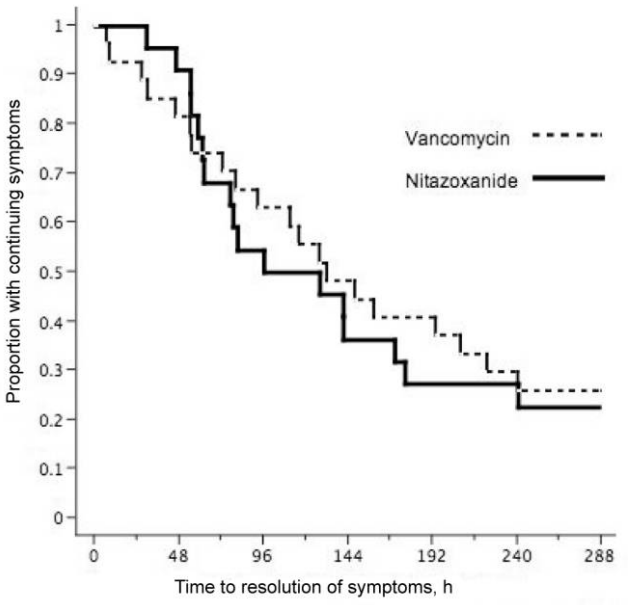

Figure 2. Kaplan-Meier analysis of time to resolution of symptoms of *Clostridium difficile* infection. $P = .55$, by Prentice-modified Wilcoxon test.

predefined noninferiority margin of −15%. The planned sample size was 175 patients per group (350 total). With 175 patients in each group, the lower limit of the observed 1-sided 97.5% CI was expected to exceed −0.15 with 86% power if the vancomycin response rate were 70% and the expected response rate for nitazoxanide were 70% (results were based on 1600 simulations) [9]. End-of-treatment response rates for vancomycin and nitazoxanide in an intent-to-treat population were each assumed to be 70% (10%–15% of patients would experience treatment failure, and 15%–20% would be unable to complete treatment because of complications of underlying illnesses).

RESULTS

Fifty patients were enrolled from November 2006 through September 2007; 27 were randomized to receive vancomycin, and 23 were to receive nitazoxanide (figure 1). The study was terminated at this point because of slower-than-expected recruitment. During treatment, 1 patient was found to have a history of inflammatory bowel disease (an exclusion criterion) and was removed from the study; when the code was revealed at study termination, he was found to be in the nitazoxanide group. Thus, the modified intention-to-treat groups consisted of 27 patients who received vancomycin and 22 who received nitazoxanide (figure 1).

The 2 groups were similar with regard to demographic characteristics and were well matched for the presence of comorbid conditions (table 1; data are shown for 49 patients), as well as for the clinical characteristics of CDI at baseline (table 2). Forty-two (86%) of 49 patients were inpatients at the start of therapy (21 [78%] of 27 vancomycin-treated and 21 [95%] of 22 nitazoxanide-treated patients; $P = .11$). Twenty (41%) of 49 patients remained in the hospital throughout the treatment period.

Four patients from each group did not complete therapy. Three patients who were receiving vancomycin and 2 patients who were receiving nitazoxanide withdrew consent because of lack of response between days 4 and 7, and 1 patient who was receiving nitazoxanide withdrew on the first day of therapy (figure 1). One patient in each group was withdrawn because of adverse events that were judged to be attributable to underlying disease and unrelated to the treatment.

A response to treatment, defined by resolution of all findings of CDI, was documented in 20 (74%) of 27 vancomycin-treated patients and 17 (77%) of 22 nitazoxanide-treated patients (difference, +3%; 95% CI, –24% to +28%). Among those who completed therapy, rates of response were 87% (20 of 23 patients) for those who received vancomycin and 94% (17 of 18 patients) for those who received nitazoxanide (difference, +7%; 95% CI, −18% to +30%). By Kaplan-Meier analysis (figure 2), time to complete resolution of all symptoms of CDI was similar in the 2 groups ($P = .55$).

After an initial response, 2 vancomycin recipients and 1 nitazoxanide recipient experienced relapse within 31 days after the start of treatment. In each of these patients, additional EIA results were positive for *C. difficile* toxin. Sustained response

Table 3. Responses to therapy in patients by severity of *Clostridium difficile* infection.

	Severe disease		Not severe disease	
Therapy result	Vancomycin group ($n = 10$)	Nitazoxanide group ($n = 10$)	Vancomycin group ($n = 17$)	Nitazoxanide group ($n = 12$)
End-of-treatment response[a]	7 (70)	8 (80)	13 (76)	9 (75)
Relapse	1 (10)	1 (10)	1 (6)	0 (0)
Sustained response	6 (60)	7 (70)	12 (71)	9 (75)

NOTE. Data are no. (%) of patients, unless otherwise indicated.

[a] Defined as complete resolution of all symptoms and signs attributable to CDI during the 3 days after completion of therapy.

rates were, therefore, 67% (18 of 27 patients) for vancomycin-treated patients and 73% (16 of 22 patients) for nitazoxanide-treated patients (difference, +6%; 95% CI, −22% to +32%) by intention-to-treat analysis; they were 78% (18 of 23 patients) for vancomycin-treated patients and 89% (16 of 18 patients) for nitazoxanide-treated patients (difference, +11%; 95% CI, −18% to +35%) by final analysis of those who completed therapy.

When patients were stratified on the basis of the severity of CDI, similar proportions in each treatment group had severe disease (table 3), and the initial and sustained responses to therapy were essentially identical in both groups. Seven patients had had CDI within 90 days before enrollment in this study. Four of the 5 vancomycin-treated patients in this group had an initial response. One of these 4 patients experienced relapse; therefore, 3 of these 5 vancomycin-treated patients had a sustained response after 31 days. Both of 2 nitazoxanide-treated patients who had had CDI within 90 days before enrollment had an initial response, and 1 experienced relapse; therefore, 1 of 2 nitazoxanide-treated patients had a sustained response after 31 days. The overall mortality rate at 31 days was 4% (2 of 49 patients).

Adverse events were common because of the prevalence of serious comorbidities. During the 31 days of observation, serious adverse events affected 4 (15%) of 27 patients who received vancomycin and 2 (9%) of 22 who received nitazoxanide; these events were all judged to be unrelated to treatment. Five vancomycin-treated patients and no nitazoxanide-treated patients had new mild-to-moderate gastrointestinal symptoms (e.g., nausea, abdominal pain, and worsening gastroesophageal reflux) during treatment that, because of the organ system that was affected, were regarded as possibly being related to therapy.

DISCUSSION

Results of this prospective, double-blind, randomized controlled trial suggest that nitazoxanide may be as effective as vancomycin in treating CDI. Eighty-six percent of patients were hospitalized at the time therapy was begun, and they had numerous underlying comorbid conditions; 41% met criteria [9] for severe disease, although patients with unstable conditions in the intensive care unit were excluded, and the overall 31-day mortality was only 4%. The rate of resolution of diarrhea and other symptoms of CDI was similar in patients treated with either drug, as was the overall success rate at 11–13 days and at 31 days after the beginning of therapy.

The strengths of this study are its double-blind, randomized design, the extent of the patients' comorbid conditions, the severity of the patients' CDI, and the fact that groups had similar demographic characteristics. These patients are similar to those commonly seen by general practitioners. The principal limitation of the study is its small size. The 50 patients enrolled provided only ~20% power to demonstrate noninferiority with use of the predefined margin of −15%. Although we did not test the antimicrobial susceptibility of *C. difficile* isolates in this study, an earlier report from the hospital that contributed the greatest number of patients to this study indicated that all isolates of *C. difficile* were susceptible to vancomycin at <8 μg/mL and to nitazoxanide at <2 μg/mL [14]; Hecht et al. [16] recently reported similar findings using strains gathered from a large number of sites in the United States.

Several alternative drugs to vancomycin, including nitazoxanide [13, 14], rifaximin [17] and OPT-80 (difimicin), are being studied for treatment of CDI [4, 11, 18]. Nitazoxanide has been widely used to treat parasitic diseases in small children and to treat cryptosporidiosis in adults [12, 19]. This drug is largely nonabsorbed from the gastrointestinal tract. It acts by inhibiting anaerobic metabolism of microorganisms. Elsewhere, we have shown this drug to be noninferior to metronidazole in treating CDI [13] and to be effective in the treatment of patients who have experienced failure of therapy with metronidazole [14]. The impact of nitazoxanide on microbial flora of the colon and its possible relationship to the emergence of bacterial resistance is unknown.

The wholesale acquisition cost of a 10-day course of nitazoxanide is $316, compared with $618 for a 10-day course of vancomycin and $5 for a 10-day course of metronidazole treatment. The cost of vancomycin can be greatly reduced by administering the intravenous preparation orally in flavored syrup. Taken together, these factors suggest that nitazoxanide can be considered for treatment of CDI with a view to reducing the use of vancomycin in hospital settings. For reasons of economy, nitazoxanide should not be substituted for metronidazole for treatment of outpatients with disease of mild-to-moderate severity. A much larger study is needed before nitazoxanide could routinely replace vancomycin in treatment of patients with severe CDI.

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