Bezlotoxumab: A Novel Agent for the Prevention of Recurrent *Clostridium difficile* Infection

Wesley D. Kufel,[1,2] (iD) Aaron S. Devanathan,[1,2] Ashley H. Marx,[1,2] David J. Weber,[3,4] and Lindsay M. Daniels[1,2]*

[1]Department of Pharmacy, University of North Carolina Medical Center, Chapel Hill, North Carolina; [2]Department of Practice Advancement and Clinical Education, University of North Carolina Eshelman School of Pharmacy, Chapel Hill, North Carolina; [3]Gillings School of Global Public Health, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina; [4]School of Medicine, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina

During the past decade, the incidence and severity of *Clostridium difficile* infection (CDI) have significantly increased, leading to a rise in CDI-associated hospitalizations, health care costs, and mortality. Although treatment options exist for CDI, recurrence is frequent following treatment. Furthermore, patients with at least one CDI recurrence are at an increased risk of developing additional recurrences. A novel approach to the prevention of recurrent CDI is the use of monoclonal antibodies directed against the toxins responsible for CDI as an adjunct to antibiotic treatment. Bezlotoxumab, a human monoclonal antibody that binds and neutralizes *C. difficile* toxin B, is the first therapeutic agent to receive United States Food and Drug Administration approval for the prevention of CDI recurrence. Clinical studies have demonstrated superior efficacy of bezlotoxumab in adults receiving antibiotic therapy for CDI compared with antibiotic therapy alone for the prevention of CDI recurrence. Bezlotoxumab was well tolerated in clinical trials, with the most common adverse effects being nausea, vomiting, fatigue, pyrexia, headache, and diarrhea. The demonstrated efficacy, safety, and characteristics of bezlotoxumab present an advance in prevention of CDI recurrence.

KEY WORDS bezlotoxumab, monoclonal antibody, *Clostridium difficile*, *Clostridium difficile* prevention, *Clostridium difficile* infection, antitoxin.

Clostridium difficile infection (CDI) is a substantial burden to patients and families and is associated with considerable morbidity and mortality.[1–3] In the United States in 2011, *C. difficile* was responsible for nearly 500,000 infections and approximately 29,000 deaths.[4] Furthermore, the direct costs of CDI in acute care settings in the United States were estimated to be $4.8 billion in 2008.[5] However, the actual costs are likely higher when indirect costs of CDI management are considered.

Currently available treatment options for initial CDI include metronidazole, oral vancomycin, and fidaxomicin. Although treatment for primary CDI is successful in many cases, CDI recurrence is common. There is at least one recurrence within the first 2 months of initial treatment with metronidazole or vancomycin in 13–47% of patients.[6–8] After the first occurrence, patients have a 38–45% chance of a second CDI recurrence, and this risk increases with each subsequent CDI recurrence.[6–8] It has been estimated that there were between 77,299 and 231,898 episodes of recurrent CDI, for approximately 560,419 to 715,018 total cases of CDI in

*Address for correspondence: Lindsay M. Daniels, Clinical Pharmacy Specialist, Infectious Diseases, Department of Pharmacy, University of North Carolina Medical Center, 101 Manning Drive, CB 7600, Chapel Hill, NC 27514; e-mail: Lindsay.Daniels@unchealth.unc.edu.

the United States in 2011.[9] Compared to primary CDI, recurrent CDI is associated with a 2.5-fold higher hospital readmission rate and a 33% higher mortality rate at 180 days, highlighting the critical need for multifaceted successful interventions.[10, 11]

The association between a healthy gut microbiota and protection against CDI has been well established in both animal and human models.[12–14] Several factors play a role in the dysbiosis of the host gut microbiota, weakening the host's natural defense system against CDI. Antibiotic use is the most important risk factor for CDI and CDI recurrence.[15, 16] Although certain antibiotics such as fluoroquinolones, clindamycin, and cephalosporins are associated with a high risk for CDI, any antibiotic that alters the normal gut microbiota can lead to CDI recurrence, including antibiotics used for CDI treatment.[17] Following a CDI treatment course, gut dysbiosis persists for a period of time until the gut microbiota reestablishes itself. *C. difficile* spores are able to germinate and reinfect the gut during this period of susceptibility after antibiotic discontinuation.[3, 18] Advanced age (> 65 yrs), prior episodes of CDI, immunosuppressing disease states, and acid-suppressing drugs (proton pump inhibitors) are also important risk factors for CDI recurrence.[19–21] Although some studies suggest no association, other studies have identified an association between the acquisition of the hypervirulent BI/NAP1/027 strain and higher rates of CDI recurrence.[22, 23]

Several nonpharmacologic interventions may reduce the incidence of health care–associated CDI with varying degrees of success. These include use of contact isolation precautions for patients with CDI, enhanced environmental disinfection, handwashing with soap and water, clinician education on appropriate *C. difficile* testing, and the implementation of antimicrobial stewardship programs to decrease the inappropriate use of antibiotic therapy.[24–29] However, these interventions have not led to a consistent or widespread decline in CDI rates.[30] Fidaxomicin is an effective treatment option for first or second CDI episodes and has been associated with a reduced rate of relapse compared to oral vancomycin, but it does not appear to be any more effective for patients with recurrent CDI.[7, 31] The use of probiotics has been controversial, where some controlled trials show benefit and others do not.[32, 33] Probiotics lack U.S. Food and Drug Administration (FDA)-approved indications for CDI, and the

term "probiotics" encompasses several different available products with varying degrees of efficacy.[33] There has also been inconsistent evidence with different adjuvant therapies such as ion-exchange resins, whole bowel irrigation, rifaximin, and nitazoxanide.[34] High cure rates for recurrent CDI have been demonstrated with fecal microbiota transplantation (FMT), but multiple factors and practical considerations may pose potential barriers to successful FMT administration.[35–37] Lack of standardized fecal delivery techniques and sources, lack of consensus on when and in what setting FMT should be pursued, third-party reimbursement considerations, and unknown utility of FMT during continued antibiotic therapy limit adoption of this modality.[37, 38] Thus, there is an urgent need for additional strategies to reduce the incidence of CDI recurrence.[38] Monoclonal antibodies actotoxumab, directed against *C. difficile* toxins A (TcdA), and bezlotoxumab, directed against toxin B (TcdB) have been pursued as therapeutic agents to prevent CDI recurrence.

Bezlotoxumab (Zinplava [formerly BLA 761046]; Merck & Co., Inc., Kenilworth, NJ), a novel humanized monoclonal antibody targeted against *Clostridium difficile* toxin B (TcdB), was approved by the FDA on October 21, 2016.[39–41] Bezlotoxumab is indicated to reduce recurrence of CDI in patients aged 18 years or older who are receiving antibacterial drug treatment for CDI and are considered at high risk for CDI recurrence.[41] Bezlotoxumab is not an antibiotic and should not be used for the treatment of CDI. This review will highlight the pharmacologic characteristics of bezlotoxumab and clinical trials for its use to prevent recurrent CDI.

Pharmacology and Mechanism of Action

Two homologous exotoxins, TcdA and TcdB, are predominantly responsible for the pathogenic effects of CDI.[42] These exotoxins lead to an increase in gut wall permeability and an acute proinflammatory response leading to diarrhea and potentially more severe symptoms of CDI.[43, 44] Targeting these toxins with serum antitoxin antibodies affords the possibility of passive immunity against recurrent CDI.[45] Evidence suggests that toxin inhibition is protective, as high titers of antitoxin antibodies correlate with lower rates of primary and recurrent CDI in humans.[46, 47] Antibody-bound toxin

is prevented from entering the gut endothelium and causing cell damage.[48, 49] Bezlotoxumab is an immunoglobulin (Ig) of the IgG$_1$ subclass, with an approximate molecular weight of 148.2 kDa.[41] Bezlotoxumab specifically binds to TcdB and neutralizes the toxin-mediated effects associated with CDI as depicted in Figure 1.

Pharmacodynamics

Bezlotoxumab binds with high affinity (K_d 19 ± 5 pmol/L) to the N-terminal half of the TcdB combined repetitive oligopeptide (CROP) domain to partially block carbohydrate binding pockets and to prevent toxin binding to host colonocytes.[50] In an in vitro study measuring the neutralization potencies of bezlotoxumab against TcdB from various *C. difficile* strains, bezlotoxumab was shown to neutralize TcdB in all strains tested.[51] In this study, although bezlotoxumab showed lower neutralization potencies for the hypervirulent strains BI/NAP1/027 and BK/NAP7/078, neutralization of these toxins was achieved at antibody concentrations below plasma concentrations seen patients with CDI. However, concentrations within the colon and feces were not specifically assessed. In another study, bezlotoxumab was shown to inhibit toxin B–mediated tumor necrosis factor α and interleukin-1β expression in human colon and peripheral blood monocytes.[52]

Pharmacokinetics

According to a population pharmacokinetic analysis, the geometric mean clearance of bezlotoxumab was 0.317 L/day with a mean volume of distribution of 7.33 L and an elimination half-life of 19 days.[53] This long half-life allows for administration of a single dose of bezlotoxumab to achieve therapeutic serum concentrations for an extended period of time to prevent recurrent CDI episodes.[53] When bezlotoxumab was administered as a single intravenous dose of 10 mg/kg, the geometric mean area under the curve from time zero extrapolated to infinity was 53,000 μg hour/ml, and the maximum serum concentration was 185 μg/ml. To our knowledge, there are no data regarding distribution of bezlotoxumab or concentrations achieved within the colonic lumen. Increased body weight was associated with an increased clearance of bezlotoxumab, and weight-based dosing accounts for increased exposure differences. Bezlotoxumab is completely eliminated by catabolism and, therefore, has a low potential for drug–drug interactions. Theoretically, no discernible drug–drug interactions are anticipated when monoclonal antibodies are coadministered, and the limited clinical observations support this prediction.[54] However, it is unknown if bezlotoxumab specifically interacts with other monoclonal antibodies. Of note, sex, race, ethnicity,
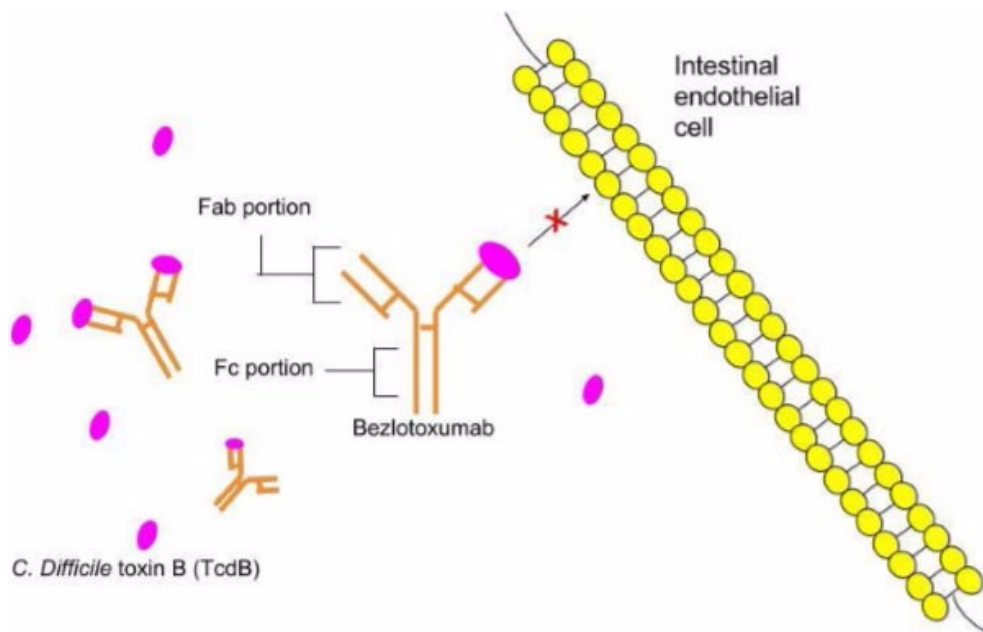

Figure 1. Bezlotoxumab binds and neutralizes *Clostridium difficile* toxin B (TcdB) to prevent TcdB from binding to the gut endothelium.

and comorbid conditions had no appreciable effect on exposure to bezlotoxumab.[41]

In patients with mild (estimated glomerular filtration rate [eGFR] 60–89 ml/min), moderate (eGFR 30–59 ml/min), or severe (eGFR 15–29 ml/min) renal impairment or with end-stage renal disease (eGFR < 15 ml/min), no clinically meaningful differences in exposure were found.[41] Similarly, hepatic impairment (defined as having at least two of the following: albumin level ≤ 3.1 g/dl; alanine aminotransferase level ≥ 2 times the upper limit of normal; total bilirubin level ≥ 1.3 times the upper limit of normal; or mild, moderate, or severe liver disease characterized by Charlson Comorbidity Index) did not yield clinically meaningful differences in exposure.[41]

Dosage, Preparation, and Administration

The recommended dosage of bezlotoxumab is a single 10-mg/kg dose administered as an intravenous infusion over 60 minutes.[41] In phase 3 clinical trials, the median patient weight in the bezlotoxumab group was 70.0 kg (range 29.8–200 kg) and, therefore, the largest dose administered was 2000 mg.[41, 55] A numerically higher number of serious adverse effects and deaths occurred in patients who weighed ≤ 70 kg compared to patients who weighed > 70 kg in all study arms. According to product labeling, the dose should be calculated based on actual body weight.[41] In clinical studies, bezlotoxumab was administered at various time points during the course of standard-of-care antibiotics for the treatment of CDI.[55] In the MODIFY I and MODIFY II trials, bezlotoxumab infusion occurred at any time point from the day prior to the initiation of antibiotic therapy to 14 days after the start of antibiotic therapy.[55] Bezlotoxumab was administered within 6 days after initiation of standard-of-care antibiotics in approximately 94% of patients, and the median administration day was on day 3.[55] Repeat administrations of bezlotoxumab have not been evaluated in clinical studies, and there is no clinical experience with bezlotoxumab overdosing.[41] Bezlotoxumab may be administered in the inpatient or outpatient clinic setting. In phase 3 clinical trials, 67.9% of participants received bezlotoxumab as inpatients.[55]

Bezlotoxumab is commercially available as a 1000-mg/40-ml (25 mg/ml) solution in a single-dose vial for intravenous use.[41] The listed average wholesale price for a 1000-mg/40-ml vial is $4560.[56] The undiluted, single-dose vial should be refrigerated (2–8°C) in the original carton to protect the product from light.[41] However, bezlotoxumab must be diluted prior to intravenous infusion once a fixed dose is selected for use. The required volume for the calculated dose should be removed from the single-dose vial and then transferred into an intravenous bag containing either 0.9% sodium chloride injection USP or 5% dextrose injection USP to prepare a diluted solution with a final concentration between 1 and 10 mg/ml.[41] The diluted product can either be stored at room temperature for up to 16 hours or under refrigeration (2–8°C) for up to 36 hours, but it should not be frozen.[41] If refrigerated, the intravenous bag needs to reach room temperature prior to administration.

Central venous access is not required for bezlotoxumab administration. It should be administered over 60 minutes using a sterile, nonpyrogenic, low-protein binding 0.2–5-μm in-line or add-on filter.[41] Bezlotoxumab should not be administered as an intravenous push or bolus, and other drugs should not be coadministered simultaneously through the same infusion line.[41]

Warnings, Precautions, and Adverse Reactions

Based on bezlotoxumab clinical trials, no contraindications currently exist.[41] However, there is a precaution for bezlotoxumab use in patients with a history of congestive heart failure (CHF), and its use should be reserved for cases where the benefits outweigh the risks in this patient population.[41] In patients with a history of CHF in phase 3 clinical trials, 12.7% (15/118 patients) of the bezlotoxumab-treated group and 4.8% (5/104 patients) of the placebo-treated group developed serious adverse reactions of CHF during the 12-week study period.[57] These adverse reactions were more common in patients with a previous, underlying diagnosis of CHF. During the 12-week follow-up period, 29% of the bezlotoxumab recipients and 33% of the placebo recipients experienced serious adverse reactions including CHF, which was reported in 2.3% of bezlotoxumab-treated patients versus 1% of placebo-treated patients. Furthermore, there were more deaths in the bezlotoxumab group (23/118 patients [19.5%]) compared to the placebo group (13/104 patients [12.5%]) in patients with a history of CHF during the 12-week study period.[55] Causes of death were various and included cardiac failure, respiratory failure, and infection. In these clinical studies, it is unclear whether the

preexisting CHF was well controlled in these patients and whether these patients had reduced or preserved ejection fraction.

The safety of bezlotoxumab was evaluated in the MODIFY I and MODIFY II trials.[55] Adverse reactions were reported within the first 4 weeks for patients who received bezlotoxumab 10 mg/kg as a single dose while on standard-of-care antibiotics for treatment of CDI. In the pooled phase 3 analysis, there were a total of 786 patients who received bezlotoxumab and 781 who received placebo. Common adverse effects were defined as occurring more frequently than placebo and reported in at least 4% of patients within the first 4 weeks of infusion. The most common adverse effects in the bezlotoxumab-treated group compared to the placebo-treated group were nausea (7% vs 5%), vomiting (3.9% vs 2.7%), fatigue (2.3% vs 1.5%), pyrexia (5% vs 3%), headache (4% vs 3%), and diarrhea (6.0% vs 5.8%).

With regard to infusion-related reactions, 10% and 8% of bezlotoxumab-treated patients and placebo-treated patients, respectively, experienced at least one infusion-specific adverse reaction on the day of or the day after administration.[55] Infusion-specific adverse reactions reported in at least 0.5% of bezlotoxumab recipients and at a frequency greater than that in placebo recipients included nausea (3%), fatigue (1%), pyrexia (1%), dizziness (1%), headache (2%), dyspnea (1%), and hypertension (1%). None of the evaluable patients in the MODIFY I and MODIFY II trials treated with bezlotoxumab tested positive for treatment-emergent anti-bezlotoxumab antibodies. During the 12-week follow-up period, overall mortality rates during this time period were 7.1% and 7.6% in the bezlotoxumab-treated patients and placebo-treated patients, respectively.[55]

No studies were performed to test the potential of carcinogenicity or impairment of fertility with bezlotoxumab administration in animals.[41] In addition, no information exists regarding the administration of bezlotoxumab during pregnancy or lactation.[41] Because the MODIFY I and MODIFY II trials only included patients aged 18 years or older, the safety and efficacy of bezlotoxumab in the pediatric population are unknown.[41]

Clinical Trial Experience

Phase 2 Clinical Trials

A phase 2, multicenter, randomized, double-blind, placebo-controlled trial evaluated the efficacy of bezlotoxumab with actoxumab (single 10-mg/kg intravenous dose of each agent) added to standard-of-care antibiotics (metronidazole or vancomycin [fidaxomicin was not FDA approved during this study period]) for the prevention of CDI recurrence.[53] The primary endpoint was the recurrence of CDI during a 12-week follow-up period, which was defined as a new episode of diarrhea associated with a new positive stool toxin test after the resolution of the initial CDI diarrheal episode and after discontinuation of metronidazole or vancomycin. Enrolled patients included those with their first episode of CDI as well as those with recurrent CDI to assess the preventive effects of these agents. Secondary outcomes measured included the severity of the initial episode, number of days to resolution of the initial episode, and antibiotic treatment failure. Diarrhea was defined as three or more unformed stools per day for at least two consecutive dates or more than six unformed stools in 1 day. The enzyme immunoassay (EIA) diagnostic testing method was used for TcdA and TcdB detection at each study site.

A total of 200 patients ($\geq$ 18 yrs of age) with diarrhea and a positive stool test for *C. difficile* toxin in the 14-day period prior to enrollment were included in the study (101 patients in the bezlotoxumab-actoxumab group and 99 patients in the placebo group). The mean age of patients was 64 years (range 20–101 yrs), and the two groups were well matched in their baseline characteristics. The rate of CDI recurrence was lower in the bezlotoxumab-actoxumab group compared to placebo (7% vs 25%, 95% confidence interval [CI] 7–29%, p<0.001). Furthermore, the relative risk (RR) of recurrence was significantly lower (RR 0.23, 95% CI 0.08–0.54, p=0.01) and the time to CDI recurrence was significantly longer (p<0.001) in the monoclonal antibody arm compared with the placebo arm.

The time to diarrhea resolution, number of days hospitalized for the initial episode, and diarrhea severity were similar between the two groups. Eighteen patients in the bezlotoxumab-actoxumab group and 28 patients in the placebo group reported at least one serious adverse effect, but this difference was not found to be statistically significant (p=0.09). The most commonly reported adverse effect was headache in both groups. Treatment with bezlotoxumab-actoxumab was effective with either concurrent metronidazole or vancomycin treatment in patients with hypervirulent strain BI/NAP1/027 or nonepidemic strains of *C. difficile*, and in

patients with their first episode of CDI or those with multiple CDI recurrences.

Data from the placebo group (n=99) were analyzed to identify risk factors associated with recurrent CDI.[58] Of the 99 patients in the placebo group, 25 patients (25.3%) had recurrent CDI within the 12-week follow-up period. The presence of serum anti-toxin B antibodies was found to be protective against CDI recurrence, but no correlation was found between serum anti–toxin A antibodies and CDI recurrence.

Phase III Clinical Trials

The safety and efficacy of bezlotoxumab in patients receiving standard-of-care antibiotics for primary or recurrent CDI were evaluated in two 12-week, phase 3, double-blind, placebo-controlled studies, MODIFY I and MODIFY II.[55] These studies included 2655 patients at 322 sites in 30 countries from November 1, 2011, through May 15, 2015, who were 18 years of age or older with a confirmed diagnosis of CDI. Efficacy was assessed in a modified intention-to-treat (mITT) group, which included all patients who received the study infusion, had a positive baseline stool test for toxigenic *C. difficile*, and began receiving standard-of-care antibiotics before or within 1 day after receiving the study infusion. Patients were included in the mITT population regardless if initial clinical cure was achieved or not after receiving standard-of-care antibiotics for CDI. Patients were excluded from these studies if surgery was required or if they had a prior diagnosis of chronic, uncontrolled diarrhea. Safety was evaluated in an as-treated population, which included all patients who received the study infusion. CDI recurrence was defined as the development of a new episode of diarrhea associated with a positive stool test for toxigenic *C. difficile* after initial clinical cure of the baseline CDI within a 12-week follow-up period.

The primary endpoint was the proportion of patients with recurrent CDI during the 12 weeks of follow-up in the mITT population following administration of bezlotoxumab alone, actoxumab alone (MODIFY I only), bezlotoxumab-actoxumab, or placebo. Initial clinical cure was defined as the absence of diarrhea for two consecutive days after the completion of standard-of-care antibiotics for ≤ 16 days. Secondary endpoints included the rate of recurrent CDI in the subgroup of patients who had initial clinical cure in the mITT population and the rate of sustained or global cure in the mITT population, which included patients with initial clinical cure and no recurrent CDI through the 12-week follow-up period. Permitted stool testing methods for diagnosis of baseline CDI included cell cytotoxicity assays, culture with toxin detection or strain typing, toxin EIA tests, or polymerase chain reaction (PCR) assays.

Patients received a single infusion of the study drug at some time point during the 10–14-day course of standard-of-care antibiotics (metronidazole, vancomycin, or fidaxomicin). Patients who received vancomycin or fidaxomicin were also eligible to receive intravenous metronidazole as combination therapy. The administration day for the study infusion as well as the choice of antibiotic therapy was at the discretion of the health care provider. The baseline characteristics were well matched across all treatment arms in both the MODIFY I and MODIFY II trials, with a median age of 65 years, 57% female, 85% white, and 68% hospitalized.

The MODIFY I trial involved 158 investigator sites in 19 countries and was funded by Merck & Co., Inc. In this study, 1396 patients were included in the mITT population: bezlotoxumab-actoxumab (383 patients); actoxumab alone (232 patients); bezlotoxumab alone (386 patients); or placebo (395 patients).[55] A total of 1224 patients completed the 12-week follow-up period. The actoxumab treatment arm was stopped early after an interim analysis demonstrated that actoxumab alone was associated with significantly higher rates of recurrent infection compared to the actoxumab-bezlotoxumab group (p=0.02). More deaths and serious adverse effects were also observed in the actoxumab group compared to the placebo group.

For the primary endpoint, the rate of recurrent CDI at week 12 was significantly lower in patients treated with bezlotoxumab alone compared with those treated with placebo, as depicted in Table 1. Initial clinical cure was achieved in 77.5% (299/386) of patients in the bezlotoxumab group compared to 82.8% (327/395) in the placebo group (adjusted difference −5.3 percentage points, 95% CI −10.9 to 0.3, p=0.0643). In patients with initial clinical cure, the differences in the rate of recurrent CDI for the bezlotoxumab group (67/299 [22.4%]) compared with the placebo group (109/327 [33.3%]) (adjusted difference −10.8 percentage points; 95% CI −17.7 to −3.8; p=0.0026) were similar to those found in the mITT population (Table 1). In addition, the proportion of patients

with sustained cure did not differ significantly between the bezlotoxumab group (232/386 [60.1%]) or the placebo group (218/395 [55.2%]) (adjusted difference 4.8 percentage points, 95% CI −2.1 to 11.7, p=0.1722).

The MODIFY II trial involved 171 investigator sites in 17 countries and was funded by Merck & Co., Inc. In this study, 1163 patients were included in the mITT population: bezlotoxumab-actoxumab (390 patients); bezlotoxumab (395 patients); or placebo (378 patients).[55] No patients received actoxumab alone due to the interim analysis performed in the MODIFY I trial. A total of 966 patients completed the 12-week follow-up period.

For the primary endpoint, the rate of recurrent CDI at week 12 was significantly lower in patients treated with bezlotoxumab alone compared to placebo, as represented in Table 1. Initial clinical cure was achieved in 82.5% (326/395) of patients in the bezlotoxumab group compared to 77.8% (294/378) of patients in the placebo group (adjusted difference 4.8 percentage points, 95% CI −0.9 to 10.4, p=0.0962). In patients with initial clinical cure, the differences in the rate of recurrent CDI for the bezlotoxumab group (62/326 [19.0%]) compared to the placebo group (97/294 [33.0%]) (adjusted difference −13.7 percentage points, 95% CI −20.4 to −6.9, p<0.001) were similar to those found in the mITT population (Table 1). Unlike the MODIFY I trial, the proportion of patients with sustained cure was significantly higher for the bezlotoxumab group (264/395 [66.8%]) compared to the placebo group (197/378 [52.1%]) (adjusted difference between bezlotoxumab and placebo 14.6 percentage points; 95% CI, 7.7 to 21.4; p<0.0001).

In an analysis of pooled data from the MODIFY I and MODIFY II trials, the rate of recurrent CDI at week 12 was significantly lower in patients treated with bezlotoxumab alone compared to placebo (Table 1).[55] Initial clinical cure

was achieved in 80.0% (625/781) of patients in the bezlotoxumab group and 80.3% (621/773) of patients in the placebo group (adjusted difference −0.3 percentage points, 95% CI −4.3 to 3.7, p=0.8832). In patients with initial clinical cure, the differences in the rate of recurrent CDI for the bezlotoxumab group (129/625 [20.6%]) compared to placebo (206/621 [33.2%]) (adjusted difference −12.2 percentage points, 95% CI −17.1 to −7.4, p<0.0001) were similar to those found in the mITT population (Table 1). Overall, the proportion of patients with sustained cure was significantly higher for patients who received bezlotoxumab alone (496/781 [63.5%]) compared with those who received placebo (415/773 [53.7%]) (adjusted difference between bezlotoxumab and placebo, 9.7 percentage points, 95% CI 4.8–14.5, p<0.0001). In the MODIFY I trial, treatment with the combination of bezlotoxumab-actoxumab did not provide added efficacy over bezlotoxumab alone.[55] This suggests the neutralization of TcdB alone to be sufficient to reduce the risk of CDI recurrence in humans. As such, the addition of actoxumab to bezlotoxumab for prevention of CDI recurrence would increase the cost of therapy and may pose additional risks.

It is important to recognize that the pooled analysis of sustained cure in the Phase III MODIFY trials does not represent a more extensive and individual analysis. In a subgroup analysis of the pooled MODIFY data, CDI recurrence rates were lower for bezlotoxumab alone compared to placebo for all subgroups evaluated.[55] Statistically significant decreases in recurrence rates were seen in patients ≥ 65 years of age, patients experiencing recurrent episodes of CDI, immunocompromised patients (based on medical history or use of immunosuppressive therapy), and patients with severe CDI. Notably, rates of recurrence in patients with the hypervirulent strain, BI/NAP1/027, were numerically, but not statistically significantly, lower in the

Table 1. Rates of *Clostridium difficile* Infection Recurrences in the Modified Intention-to-Treat Analysis of Phase III Clinical Trials of Bezlotoxumab

| Trial | *Clostridium difficile* Infection Recurrence Rate at Week 12 | | |
	No. (%) of Patients Receiving Bezlotoxumab	No. (%) of Patients Receiving Placebo	Absolute Difference in *Clostridium difficile* Infection Recurrence Rate (Bezlotoxumab − Placebo)
MODIFY I (phase 3)[55]	67/386 (17.4)	109/395 (27.6)	−10.1% (95% CI −15.9 to −4.3), p=0.0003
MODIFY II (phase 3)[55]	62/395 (15.7)	97/378 (25.7)	−9.9% (95% CI −15.5 to −4.3), p=0.0003
MODIFY I and MODIFY II pooled data[55]	129/781 (16.5)	206/773 (26.6)	−10.0% (95% CI −14.0 to −6.0), p<0.0001

CI = confidence interval.

bezlotoxumab group (21/89 [23.6%]) compared to the placebo group (34/100 [34.0%]).[55] However, a statistically significant decrease in recurrence was seen for patients with the BI/NAP1/ 027 strain who received the combination of bezlotoxumab plus actoxumab (9/76 [11.8%]) compared to placebo (34/100 [34%]). This finding suggests that the addition of actoxumab to bezlotoxumab may provide a greater benefit for patients with the BI/NAP1/027 strain compared to bezlotoxumab alone.

Based on the pooled data analysis and absolute risk reduction (ARR) data presented in Table 1, the number needed to treat (NNT) to prevent one CDI recurrence episode was 10.[55] However, the NNT was six among patients with the identified risk factors of age ≥ 65 years (15.4% had CDI recurrence with bezlotoxumab compared to 31.4% with placebo) and those with at least one previous CDI in the past 6 months (25.0% had CDI recurrence with bezlotoxumab compared to 41.1% with placebo).[55] At 6-, 9-, and 12-month follow-up periods, *C. difficile* colonization was similar in all study groups, ranging from 16.3% to 32.8%.[59] This suggests that the efficacy of bezlotoxumab observed during the 12-week follow-up period was attributable to sustained protection from CDI recurrence rather than a delay in the onset of a recurrent CDI.[59] In addition, most CDI recurrences (71%) occurred within 4 weeks after study infusion.[55]

Several different testing methods for diagnosis of CDI were used in the MODIFY trials. A post hoc pooled analysis was performed to examine the potential impact of EIA versus PCR diagnostic testing on study outcomes since each method varies in its predictive value for true positive CDI.[60] Stool testing methods for diagnosis of baseline CDI included cell cytotoxicity assays, culture with toxin detection or strain typing, toxin EIA, or PCR. All permitted commercial testing methods had the capacity to detect the presence of TcdB (EIA method) or the TcdB gene (PCR method) and had a labeled specificity of $\geq 94\%$. The EIA testing method was the most commonly used diagnostic test at baseline, followed by PCR, culture with toxin detection or strain typing, and cell cytotoxicity assay. CDI recurrence rates were higher when diagnosis was made by PCR compared with toxin EIA for bezlotoxumab-treated patients. In contrast, CDI recurrence rates were similar for diagnoses using PCR or EIA for placebo-treated patients. The reduction of CDI recurrence associated with bezlotoxumab was larger if EIA rather than PCR was used to diagnose CDI. CDI recurrence rates among patients diagnosed with EIA were 27.3% (105/385 patients) in the placebo group versus 14.5% (54/372 patients) in the bezlotoxumab group. For patients diagnosed with PCR, the recurrence rates were 26.1% (88/337 patients) in the placebo group versus 19.6 (70/357 patients) in the bezlotoxumab group.[60]

Practical Considerations and Future Directions

Although the MODIFY I and MODIFY II clinical trials provided important evidence to support the role of bezlotoxumab in the prevention of CDI recurrence and led to FDA approval for this agent, several important questions remain. The duration of benefit after administration of bezlotoxumab beyond the 12-week follow-up period in these trials is unknown. Furthermore, it is unknown which patient populations are likely to derive the greatest benefit from bezlotoxumab administration. Bezlotoxumab is indicated for patients who are considered high risk for CDI recurrence, yet these patient populations are not well defined within the product labeling. Patients at high risk for recurrence may include older adults, immunosuppressed hosts, patients with a history of CDI recurrence, and those receiving antibiotic therapy.[17] The role of bezlotoxumab in primary CDI prevention for high-risk patient groups is another interesting area to consider for future research in order to determine if this is both clinically and economically justifiable.

It is also unclear how CDI recurrence rates compare between patients who receive bezlotoxumab and those who receive alternative preventative therapies such as FMT or other novel approaches in the pipeline for prevention of CDI. FMT has been demonstrated to be highly effective in treating CDI as well as reducing CDI recurrence.[38] Although head-to-head trials comparing clinical efficacy and safety of FMT to bezlotoxumab for prevention of CDI recurrence could be useful, it is important to highlight that FMT is a treatment modality whereas bezlotoxumab is only a preventive agent that must be used in conjunction with CDI treatment agents. A total of 26 patients (22 in the placebo group, 4 in the bezlotoxumab group) received FMT in the MODIFY I and II trials, but their outcomes are not available. Bezlotoxumab may offer an advantage over FMT in individualized patient-specific situations, particularly in patients who

do not respond to FMT or those who are unable to stop antibiotic therapy for other concurrent infectious diseases indications.

The degree of benefit observed for each specific standard-of-care antibiotic (metronidazole, vancomycin, or fidaxomicin) in combination with bezlotoxumab remains unclear as well since the MODIFY trials did not provide relative risk stratification for different antibiotics used with bezlotoxumab.[38, 55] In the pooled analysis of the MODIFY I and MODIFY II trials, only 4% of patients received fidaxomicin.[55] Fidaxomicin alone has been associated with a reduction in CDI recurrence rates among patients treated for their first CDI episode for the nonvirulent, non-NAP1/BI/027 strains.[7] The additional benefit of bezlotoxumab in patients treated with fidaxomicin remains unknown.[7, 38] It is also important to highlight that the follow-up period to assess for CDI recurrence was much shorter in phase 3 clinical trials for fidaxomicin compared to bezlotoxumab (28 days for fidaxomicin versus 84 days for bezlotoxumab).[7, 55] The phase 3 clinical trials that led to the FDA approval of fidaxomicin excluded patients with more than one episode of CDI in the previous 3 months, which may also influence CDI recurrence rates for patients with multiple CDI recurrences, as these patients have exposure to multiple antibiotic courses for CDI treatment and an altered gut microbiota.[7]

In the pooled data analysis of the MODIFY I and MODIFY II trials, the NNT to prevent one episode of CDI recurrence was 10.[55] In clinical practice, the actual NNT may differ from this, as the approach to bezlotoxumab use remains to be determined. In the MODIFY trials, bezlotoxumab was given to patients being treated for CDI regardless of risk factors and regardless of whether it was a first or recurrent episode. It is also important to note that there was a difference in the CDI recurrence rates for different diagnostic testing methods, with higher rates of CDI recurrence observed when PCR was used for diagnosis compared to EIA testing. Thus, the NNT to prevent recurrence will be higher than that seen in the clinical trials for populations in which PCR is the primary diagnostic test used. This is an important aspect to consider when making formulary decisions, particularly for institutions that solely use PCR diagnostic testing.

Prior to the widespread adoption of bezlotoxumab in health care settings, a cost-benefit analysis needs to be performed to best determine its role in therapy and to determine whether this intervention is economically beneficial for health care institutions and specific health care settings. With an approximate 10% ARR presented in phase 3 clinical trials, it will be critical for institutions to determine whether bezlotoxumab is economically viable and clinically justifiable based on their specific institution, patient populations, and health care–related costs regarding CDI recurrence. Careful patient selection and risk stratification may translate into a more cost-effective approach to decrease CDI recurrence rates. Optimal timing of administration will also need to be determined for various patient populations at high risk for relapse. Third party reimbursement, patient location, and urgency of administration will likely play a role in whether bezlotoxumab is administered in an outpatient clinic setting versus an inpatient setting. Bezlotoxumab was administered at various time points during standard-of-care antibiotics for CDI treatment. Thus, bezlotoxumab administration potentially could be deferred to the outpatient setting once patients improve clinically to minimize inpatient-related costs. Based on these considerations, there are potential concerns and unresolved issues that should be addressed in future studies to better characterize the role of bezlotoxumab in the clinical setting.

Conclusion

Bezlotoxumab, a fully humanized monoclonal antibody that binds to and neutralizes *C. difficile* toxin B, is the first-in-class, FDA-approved agent to provide passive immunity for the prevention of CDI recurrence. In clinical studies, bezlotoxumab was well tolerated and effective for the reduction of CDI recurrence compared to placebo. Pharmacoeconomic analyses are needed to guide cost-effective use. Although much of the role of bezlotoxumab remains to be revealed by phase 4 clinical experience, this agent is a welcomed addition to the CDI management armamentarium, where limited therapeutic options exist.

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