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RBX2660

SPONSOR BRIEFING DOCUMENT

VACCINES AND RELATED BIOLOGICAL PRODUCTS ADVISORY COMMITTEE

MEETING DATE: 22 SEPTEMBER 2022

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

Abbreviation	Definition
AE	Adverse event
AESI	Adverse events of special interest
ATLAS score	Age, Temperature, Leucocytes, Albumin, and Systematic antibiotics score
BLA	Biologics License Application
C. difficile	*Clostridioides difficile*
CCI	Charlson Comorbidity Index
CDAD	*C. difficile*-associated diarrhea
CDBPCR	*Clostridioides difficile* Toxin B polymerase chain reaction
CDC	Centers for Disease Control and Prevention
CDI	*Clostridioides difficile* infection
CFU	Colony-forming units
cGMP	Current Good Manufacturing Practice
CI	Confidence interval
CKD	Chronic kidney disease
CNS	Central Nervous System
CRE	Carbapenem-resistant *Enterobacteriaceae*
CrI	Credible Interval
DHS	Donor human stool
DSMB	Data and Safety Monitoring Board
EAC	Endpoint adjudication committee
EIA	Enzyme-linked immunosorbent assay
EPEC	Enteropathogenic *Escherichia coli*
ESBL	Extended-spectrum beta-lactamase-producing *Enterobacteriaceae*
FDA	Food and Drug Administration
FMT	Fecal microbiota transplantation
GDH	Glutamate dehydrogenase
GI	Gastrointestinal
GMP	Good Manufacturing Practice
HIPAA	Health Insurance Portability and Accountability Act
HIV	Human immunodeficiency virus
HTCP	Human Cell and Tissue Products
IBD	Inflammatory bowel disease
IBS	Irritable bowel syndrome
ICH	International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use
ICU	Intensive Care Unit
IND	Investigational New Drug
ISS	Integrated summary of safety
ITT	Intent-to-treat
MDRO	Multi-drug resistant organism
MedDRA	Medical Dictionary for Regulatory Activities
mITT	Modified intent-to-treat
MRSA	Methicillin-resistant *Staphylococcus aureus*
MRT	Microbiota restoration therapy
PCR	Polymerase chain reaction
PEG	Polyethylene glycol 3350
PI	Principal investigator
PP	Per protocol
rCDI	Recurrent *Clostridioides difficile* infection
SAE	Serious adverse event

Abbreviation	Definition
SD	Standard deviation
SMQ	Standardized MedDRA Query
SoC	Standard of care
SOC	Standard of care
SP	Safety Population
STEC	Shigella-toxin-producing *Escherichia coli*
US	United States
UTI	Urinary tract infection
VRE	Vancomycin-resistant *Enterococci*
VRE	Vancomycin-resistant *Enterococcus*

1 EXECUTIVE SUMMARY

1.1 Introduction

Clostridioides difficile (*C. difficile*) is an anaerobic bacterial pathogen that produces toxins that result in inflammation of the intestines, causing diarrhea, and more severely, colitis and sepsis. *C. difficile* infection (CDI) is a serious and potentially life-threatening disease and is responsible for considerable morbidity and mortality. The Centers for Disease Control and Prevention (CDC) lists CDI as an Urgent Threat, caused more than 18,000 deaths in a recent year and is a leading Healthcare Acquired Infection (HAI) (CDC, 2019). Following a course of antibiotic therapy for CDI, 20% to 30% of patients will experience recurrent CDI (rCDI) (Sheitoyan-Pesant et al., 2016), which requires further antibiotic therapy and can lead to a cycle of additional recurrences. Once the disease has recurred, the risk of further recurrences is increased, with 40% to 60% of patients experiencing additional recurrent episodes (C. P. Kelly, 2012; Wenisch, Parschalk, Hasenhundl, Hirschl, & Graninger, 1996).

RBX2660 is an investigational fecal microbiota-based live biotherapeutic developed to reduce recurrence of CDI in adults following antibiotic treatment for rCDI. It is a pre-packaged, single-dose, 150 mL fecal microbiota suspension that is rectally administered. During development, RBX2660 was granted Fast Track, Breakthrough Therapy, and Orphan Drug designations from the Food and Drug Administration.

From an efficacy standpoint, RBX2660 demonstrated a statistically significant and clinically meaningful reduction in rCDI in the pivotal Phase 3 trial. This result is further supported by consistently favorable and sustained treatment responses observed throughout the entire clinical development program and builds upon the well-recognized concepts of fecal microbiota transplant, which, while unapproved, has been used for decades and is now included in clinical guidelines.

With regard to safety, RBX2660 was well tolerated in randomized, placebo-controlled trials, with mild to moderate gastrointestinal events, such as abdominal pain and diarrhea, being the most frequently reported. This safety profile was consistent across the clinical program.

1.2 Background and Unmet Need

C. difficile is the most common pathogen causative of healthcare-associated infections in the United States (US), causing almost half a million infections each year, tens of thousands of deaths and has been declared an urgent antibiotic resistance threat and an urgent national health threat by the CDC (CDC, 2015, 2020a, 2020b; Lessa et al., 2015).

Recurrence of CDI is a significant health problem and there are currently few solutions available to help people who experience rCDI. The risks of rCDI increase with each subsequent recurrence. The community of microorganisms resident in the human intestinal tract, or gut microbiome, is recognized as a key regulator of metabolic and immune homeostasis and a mediator of resistance to some pathogenic infections, including *C. difficile*. In some cases, antibiotic treatment can disrupt the composition and diversity of the gut microbiome, decreasing its resistance to *C. difficile* colonization, which in turn can allow *C. difficile* to proliferate and produce toxins that cause destruction of colonic epithelial cells, inflammation, and disease symptoms. Antibiotic treatment is the current standard of care for treating CDI but can result in continued gut microbiome

disruption, allowing *C. difficile* to recolonize and CDI to recur. To help patients break this cycle of recurrence, alternatives to antibiotics are needed to reduce the risk of rCDI.

There are limited approved non-antibiotic treatments currently available for rCDI.

Bezlotoxumab is a human monoclonal antibody that was studied in patients with primary CDI and rCDI, and is approved for marketing in the US. Bezlotoxumab binds to *C. difficile* Toxin B, one of two toxins produced by the bacterium, and is indicated to reduce rCDI in patients 18 years of age or older who are receiving antibacterial drug treatment for CDI and are at a high risk for CDI recurrence (Merck & Co., 2016). Bezlotoxumab carries the risk of heart failure, especially in patients with a history of congestive heart failure and should be reserved for patients in whom the benefits outweigh the risks (Merck & Co., 2016).

Fecal microbiota transplantation (FMT has demonstrated efficacy in reducing rCDI and is thought to restore the composition and diversity of the gut microbiome after antibiotic treatment, thereby suppressing *C. difficile* colonization and outgrowth. Though not studied in a large well-controlled setting, in general, the effect of FMT as an effective treatment option after second recurrence of rCDI is considered well-established following decades of use in the clinical setting. However, it remains an unapproved therapeutic procedure with no standardized formulation, strength, application, or administration.

An FDA approved, standardized, and accessible treatment to reduce the recurrence of CDI by restoring the gut microbiome would give patients and health care providers a new option for this life-threatening infection. Availability of a well-studied product with an established and consistent benefit-risk profile would reduce variability and heterogeneity compared with the current wide range of FMT processes and preparations. An FDA approved microbiota restoration product would improve access for patients to treatment that effectively reduces the cycle of CDI recurrence.

1.3 Product Description

RBX2660 is a fecal microbiota suspension prepared from donor human stool (DHS) which is collected from pre-screened and qualified donors. In order to ensure consistency and quality, the suspension is tested and processed under strict Good Manufacturing Practice (GMP) conditions to provide a stable, cryopreserved drug product. Rebiotix has implemented donor qualification and pathogen screening processes since its initial IND application in 2012. During this time, the program has been reviewed, updated and validated under FDA oversight. This process employs comprehensive testing of each donor and each donation for a broad panel of infectious agents to ensure patient safety. RBX2660 drug product is supplied as a pre-packaged single dose 150 mL fecal microbiota suspension containing a consortia of $1x10^8$ to $5x10^{10}$ colony-forming units (CFU)/mL of diverse viable bacteria, including Bacteroides, which have been linked to resistance to *C. difficile* colonization.

1.4 Development Program

The clinical development program for RBX2660 included 6 studies:

- Two randomized placebo-controlled studies (Phase 2B Study 2014-01 and Pivotal Phase 3 Study 2017-01);

- Three open-label studies (Phase 2 Studies 2013-001 and 2015-01, and the ongoing Phase 3 Study 2019-01); and
- One retrospective study (Study 2019-02).

All of these studies, placebo-controlled and open-labeled, contribute to the overall totality of evidence supporting the safety and efficacy of RBX2660. Study 2014-01 supported the selection of 1 dose of RBX2660 as the treatment regimen used in Study 2017-01. The results of the Pivotal Phase 3 Study 2017-01 constitute the primary evidence of efficacy.

The nonrandomized, open-label studies (Studies 2013-001, 2015-01, 2019-01, and 2019-02) provide supplementary evidence of efficacy and safety for RBX2660 for its proposed indication. Studies 2019-01 and 2019-02 enrolled a more diverse patient population, including some patients with irritable bowel syndrome (IBS), inflammatory bowel disease (IBD), and immunocompromised conditions, who could not have qualified for study enrollment in Studies 2017-01 and 2014-01.

1.5 Program Context

The development program was originally planned to include two randomized placebo-controlled pivotal Phase 3 trials. However, during the development program, the widespread availability of unapproved FMT under the 2013 FDA guidance on Enforcement Discretion made it increasingly difficult to enroll patients within this orphan indication. In addition, rCDI treatment guidelines were updated to include recommendations to consider FMT therapy, acknowledging its experimental and unapproved status. For a debilitating and life-threatening disease such as rCDI, availability of a treatment that is similar to the investigational product may dissuade patients from enrolling in a trial in which they may be randomized to placebo treatment. As a consequence, the availability of patients meeting the enrollment criteria and willing to participate in placebo-controlled trials steadily declined over time.

FDA acknowledged the increasing recruitment difficulties and recommended that innovative design options, such as formal borrowing of data in a Bayesian framework, could be pursued to reduce the number of patients required to demonstrate evidence of effectiveness in a single pivotal trial. The Bayesian approach was selected, because it is ideally suited for incorporating multiple sources of information together in a single analysis. Therefore, Study 2017-01 was analyzed using a Bayesian hierarchical model that was developed in discussions with the FDA. This analysis method integrates the data from the Phase 2B Study 2014-01 and the Phase 3 Study 2017-01 into a single analysis estimating the relative efficacy of the RBX2660 treatment compared to placebo in the Phase 3 study.

Additional information about the statistical analysis for the primary efficacy endpoint of the Pivotal Phase 3 Study is provided in Section 6.4.1.5.2 and a detailed report on the adaptive design is included in Appendix 11.1.

1.6 Efficacy Findings

1.6.1 Phase 2B Study 2014-01 Results

Study 2014-01 was a Phase 2B, prospective, multicenter, randomized, double-blind, placebo-controlled, regimen-finding study, evaluating the efficacy and safety of RBX2660 in adults with 2 or more episodes of rCDI or at least 2 episodes of severe CDI resulting in hospitalization. A total of 150 patients were enrolled. Patients were required to have a positive stool test for the presence

of *C. difficile* within 60 days prior to enrollment. Patients were randomized 1:1:1 to evaluate 3 different treatment regimens with the doses for all groups administered 1 week apart:

- 2 doses of RBX2660,
- 1 dose of RBX2660 followed by 1 dose of placebo, and
- 2 doses of placebo.

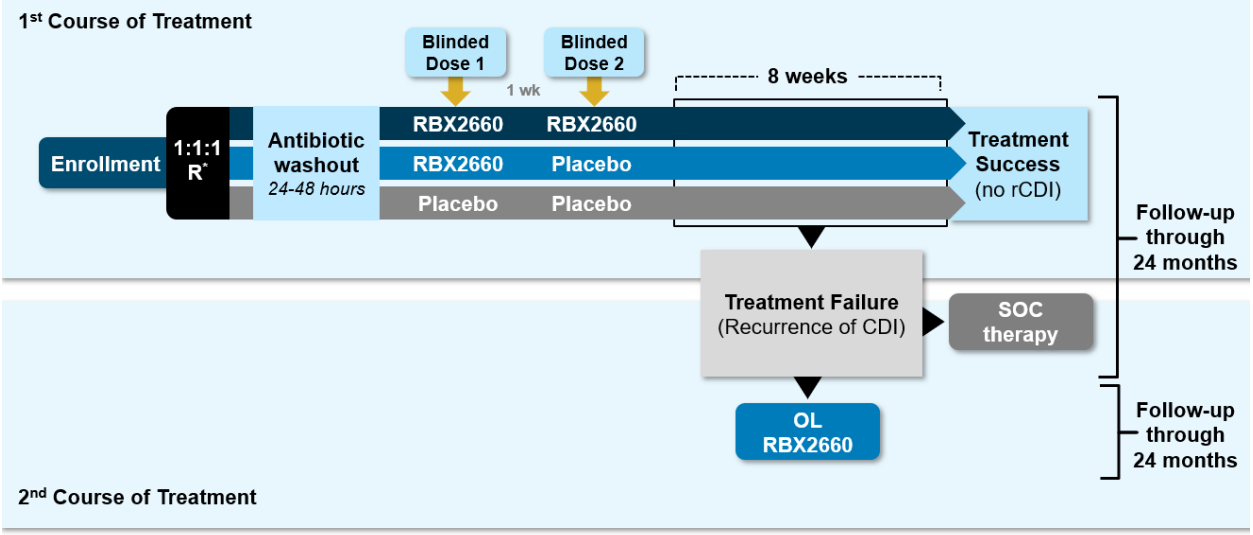
Patients who had a confirmed recurrence of CDI within 8 weeks were offered an open-label treatment course of RBX2660 (Figure 1).

The primary efficacy endpoint was Treatment Success, defined as the absence of *C. difficile*-associated diarrhea for 8 weeks after administration of the last study treatment. The primary efficacy analysis was performed using the Intent-to-Treat (ITT) population, which consisted of all randomized patients, regardless of whether they completed their assigned study treatment. A total of 133 patients (45 in the 2-Dose RBX2660 Group, 44 in the 1-Dose RBX2660 Group, and 44 in the Placebo Group) were included in the ITT population.

In the final analysis of primary efficacy, for the comparison of the 2-Dose RBX2660 Group (N=25/45; 55.6% Treatment Success) to the Placebo group (N=19/44; 43.2% Treatment Success) in the blinded portion of the study, the statistical significance of the primary efficacy endpoint was not met (p=0.243).

Regarding treatment regimen selection for the subsequent clinical studies, an analysis comparing the 2 RBX2660 treatment regimens showed no meaningful difference in efficacy between treatment with 1 dose (Treatment Success, 56.8% [25/44]) versus 2 doses administered 1 week apart (Treatment Success, 55.6% [25/45]); therefore, a single dose of RBX2660 was selected for the Pivotal Phase 3 Study.

Figure 1: Phase 2B Study 2014-01 Overall Design



*Stratified at baseline by type of antibiotic used: vancomycin, fidaxomicin, other; RBX2660/RBX2660 = 2 doses, RBX2660/Placebo = 1 dose; SOC: standard of care

1.6.2 Pivotal Phase 3 Study 2017-01 Results

Study 2017-01 was a prospective, multicenter, randomized, double-blinded, placebo-controlled Phase 3 study to confirm the efficacy and safety of RBX2660 for the prevention of rCDI in patients

who have had prior rCDI that was resolved with antibiotic treatment. Randomization was at a 2:1 ratio of 1 dose of RBX2660 to placebo, with a target enrollment of 270; approximately 180 patients with a positive stool test for the presence of toxigenic *C. difficile* within 30 days prior to enrollment were to be randomized to RBX2660 treatment and 90 patients randomized to placebo (Figure 2).

The primary efficacy endpoint for Study 2017-01 was the absence of CDI diarrhea for 8 weeks after study treatment. The primary efficacy analysis was performed using the mITT Population, which included all randomized patients who successfully received blinded treatment, excluding those who discontinued from the study for reasons not related to CDI symptoms prior to evaluation of Treatment Success for the primary endpoint. A total of 262 patients (177 in the RBX2660 group and 85 in the Placebo group) were included in the mITT population.

The efficacy analysis of the primary efficacy endpoint used a Bayesian hierarchical model, which was developed in discussions with the FDA and implemented prior to enrollment completion, data unblinding, and analyses of study data. The Bayesian design incorporated both dynamic borrowing of information from the prior randomized placebo-controlled Phase 2 study (Study 2014-01), as well as 2 interim analyses of the 2017-01 study performed to determine whether the trial should stop early for futility or overwhelming evidence of efficacy. The study included 2 success criteria levels at the final analysis that were adjusted for early efficacy stopping Type I error spending:

- A posterior probability of superiority greater than 97.50% but less than 99.93%, which would be considered equivalent to a single positive adequate and well-controlled trial; and
- A posterior probability of superiority greater than 99.93%, a statistically very persuasive finding in a single trial that would be considered as providing equivalent statistical evidence to 2 positive adequate and well-controlled trials.

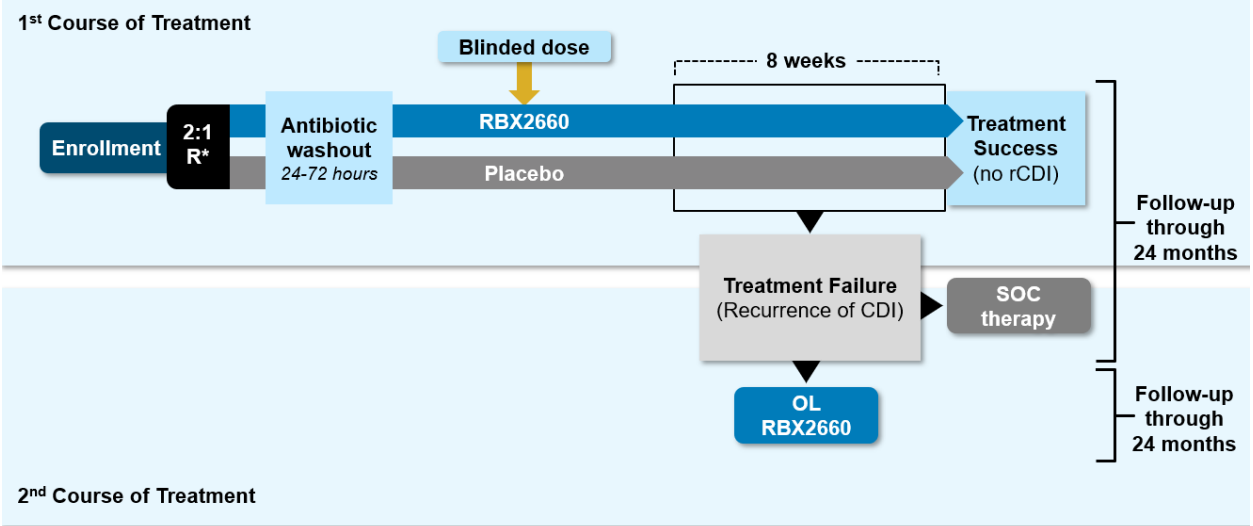
At the final planned analysis, RBX2660 was superior to placebo in the prevention of CDI recurrence through 8 weeks. The model-estimated Treatment Success rate was 70.4% in the RBX2660 group and 58.1% in the Placebo group. The difference in Treatment Success rates between the RBX2660 and Placebo groups was 12.3 percentage points (95% credible interval [CrI]: 1.4 to 23.3) with a 98.6% posterior probability that RBX2660 was superior to placebo. Thus, the study met the first success criterion but did not meet the higher success criterion.

However, during Biologics License Application (BLA) review, FDA pointed out that aligning the analysis populations and definitions would lead to a stronger claim to exchangeability between Studies 2014-01 and 2017-01. Therefore, an updated primary efficacy endpoint analysis was performed using the Bayesian hierarchical model, applying the Study 2017-01 definitions of the analysis populations to the Study 2014-01 efficacy data, matching populations when borrowing, and restricting the follow-up period in Study 2014-01 to 8 weeks from first dose. The model-estimated Treatment Success rate was 70.6% in the RBX2660 group and 57.5% in the Placebo group. The difference in Treatment Success rates between the RBX2660 and Placebo groups was 13.1 percentage points (95% CrI: 2.3 to 24.0) with a 99.1% posterior probability that RBX2660 was superior to placebo.

Sensitivity analyses of the primary endpoint using the Bayesian hierarchical model in the ITT and Per Protocol (PP) populations showed results consistent with the primary analysis. Results of Treatment Success in subgroups were generally consistent with the primary analysis, including analyses by age, sex, and race. See Section 6.4.3 for additional details on analysis results.

Patients who had Treatment Failure within the first 8 weeks (whether they received RBX2660 or placebo) were offered a second treatment with open-label RBX2660. Among the 51 patients who had been treated with RBX2660 during the blinded period of the study but had Treatment Failure, 41 patients were eligible and chose to receive a second treatment with open-label RBX2660, and more than half (22/41; 53.7%) had absence of CDI recurrence within 8 weeks of completing treatment. This result, combined with the rate of success after 1 course of treatment with RBX2660, showed an overall rate of approximately 83.6% Treatment Success after patients received up to 2 courses of RBX2660 treatment.

Figure 2: Pivotal Phase 3 Study 2017-01 Overall Design



*Stratified at baseline by type of antibiotic used: vancomycin, vancomycin in combination, fidaxomicin, other; SOC: standard of care

1.7 Safety Findings

Safety data from the Pivotal Phase 3 Study 2017-01, as well as integrated safety analyses of data from across the 6 studies of RBX2660 (2 placebo-controlled studies, 3 open-label studies, and 1 retrospective study, with a total of 978 patients exposed to RBX2660) show that the safety profile was consistent across the clinical program.

Regarding safety in Phase 3 Study 2017-01 during the initial 8-week double-blind period:

- AEs were reported for 47.8% (86/180) of patients in the RBX2660 and 39.1% (34/87) of patients in the Placebo group, and most AEs in patients treated with RBX2660 were mild or moderate in severity.
    - The most common AEs were gastrointestinal, with the most common being diarrhea (12.2% [22/180] in the RBX2660 group and 12.6% [11/87] in the Placebo group) and abdominal pain (12.8% [23/180] in the RBX2660 group and 10.3% [9/87] in the Placebo group). The gastrointestinal AEs typically occurred early (within the first 7 days of starting treatment), and were short in duration, lasting a median of 2 days.
- SAEs were reported for 2.2% (4/180) of patients in the RBX2660 group and 1.1% (1/87) of patients in the Placebo group.

- One death was reported during the double-blind period of Study 2017-01; the death was from cardio-respiratory arrest, in a patient with extensive cardiovascular medical history.

The integrated safety population included all patients who received at least 1 dose of study treatment in the three Phase 2 studies and two Phase 3 studies. Because the integrated database combines open-label and blinded study data, there is a large imbalance in the number of patients exposed to RBX2660 (N=978) as compared to placebo (N=83). The analyses in the integrated dataset were primarily of AEs which had onset within 6 months of last treatment. Regarding safety findings in this integrated population:

- AEs were reported for 68.8% (673/978) of patients in the All RBX2660 group and most AEs were mild or moderate in severity.
- The most common AEs were gastrointestinal in nature, with the most common being:
    - Diarrhea: 23.1% (226/978).
    - Abdominal pain: 16.4% (160/978).
    - Nausea: 9.3% (91/978).
- SAEs were reported for 13.8% (135/978) of patients in the All RBX2660 group, with the most common SAE by preferred term being *C. difficile* infection, with a frequency of 2.6% in the All RBX2660 group, and was due to recurrences of CDI meeting SAE criteria. *C. difficile* infection was the only term with >1% frequency, and the remaining events showed no signs of clustering by system organ class.
- Across all studies, there were 18 AEs with onset within 6 months of last treatment with RBX2660 leading to patient death. There were no deaths among patients given placebo from AEs with onset within 6 months of blinded treatment. However, the exposure time to placebo was 42 patient-years in total while exposure time for the All RBX2660 group was 404 patient-years. A summary of AEs leading to patient death by observation time is provided in Table 25. The types of events leading to death were a variety of preferred terms and system organ classes with no pattern or clustering of AEs.

Overall, RBX2660 was well tolerated, with generally expected and manageable AEs.

1.8 Benefit-Risk Summary

RBX2660 shows a clinically meaningful benefit for patients suffering from rCDI. The statistically significant results of the pivotal, placebo-controlled, Phase 3 study are supported by a totality of evidence of both strong benefits and defined risks of treatment from the open-label studies. The reduction of recurrences of CDI through 8 weeks was durable. Breaking the cycle of CDI recurrence has clear benefits, given the considerable morbidity and mortality of the condition.

RBX2660 is well tolerated, with the risks both known and manageable. The risks of RBX2660 include diarrhea and abdominal pain, which were the most common AEs and were predominately mild to moderate.

The benefit of RBX2660 outweighs these risks while addressing an urgent unmet medical need by providing a novel treatment option intended to restore the microbiome and reduce rCDI. Physicians and patients need a well-studied, approved restorative therapy that has an established

and favorable benefit-risk profile. Approval of a product that is developed, manufactured, tested and regulated under FDA oversight would improve access for patients.

2 BACKGROUND ON *CLOSTRIDIOIDES DIFFICILE* INFECTION (CDI)

Summary

- *Clostridioides difficile* (*C. difficile*) is an anaerobic bacterial pathogen that can produce toxins resulting in inflammation of the intestines, causing diarrhea, and more severely, colitis and sepsis.
- *C. difficile* infection (CDI) is a serious and potentially life-threatening illness that is responsible for considerable morbidity, mortality, and healthcare expenditures.
- Antibiotic treatment is the current standard of care for treating CDI, but recurrence after cessation of antibiotics is common, occurring in around 20% to 30% of patients (Sheitoyan-Pesant et al., 2016), which requires further antibiotic therapy. Thus, antibiotic treatment for CDI may initiate a cycle of recurrence caused by the dysbiosis introduced by antibiotic treatment.
- Fecal microbiota transplantation (FMT), which is thought to restore the composition and diversity of the gut microbiome and thereby suppress *C. difficile* outgrowth, is commonly used as a treatment for rCDI (Khanna, 2021), though it is not currently approved in the US.
- Development of microbiome-based therapeutics for patients with rCDI is urgently needed to fulfill an important unmet medical need.

2.1 Overview of *Clostridioides difficile* Infection (CDI)

2.1.1 Overview of Disease

Clostridioides difficile (*C. difficile*) is an anaerobic bacterial pathogen that produces toxins that can induce inflammation of the intestines, causing diarrhea, and in severe cases, colitis, sepsis and death. *C. difficile* is a leading HAI in the US and has been declared an urgent antibiotic resistance and Urgent Threat by the Centers for Disease Control and Prevention (CDC) (CDC, 2019, 2020b). *C. difficile* causes almost half a million infections in the US each year, with up to 20-30% or more patients experiencing recurrence (CDC, 2020a; Johnson et al., 2021; C. P. Kelly, 2012; Lessa et al., 2015; Sheitoyan-Pesant et al., 2016; Smits, Lyras, Lacy, Wilcox, & Kuijper, 2016). *C. difficile* infection is a serious and potentially life-threatening illness and is responsible for considerable morbidity, mortality, and healthcare expenditures.

In 2011, CDI incidence and subsequent thirty-day all-cause mortality rates were estimated to range from 1.3 to 9.3%, depending on whether the infection was community-acquired or healthcare-associated (Lessa et al., 2015). Mortality rates are particularly high for patients over age 65 diagnosed with healthcare-associated CDI; the 30-day mortality estimate for patients aged 65 and older was 9.1% (all-cause, after first CDI) (CDC, 2020a). CDI has become an epidemic and continues to gain momentum, with an escalation in overall incidence and severity of disease in recent decades (Bakken et al., 2011; Cohen et al., 2010; Czepiel et al., 2019). Mortality rates have appeared to remain stable over time (from 2011 to 2017) despite a decrease in the burden of hospitalizations among healthcare-associated CDIs (Guh et al., 2020). Patients with CDI also deal with a substantial burden of physical and psychological effects, with significant reduction in quality of life during CDI and after recovery from CDI (Lurienne et al., 2020).

Recurrent CDI (rCDI) is defined as an episode of CDI occurring within 8 weeks of a previous episode (C. R. Kelly et al., 2021a), and is associated with significant increased mortality. Patients with rCDI had 33% higher risks of death at 180 days compared with patients without rCDI (Olsen, Yan, Reske, Zilberberg, & Dubberke, 2015).

2.2 Current Treatment Options/Paradigm

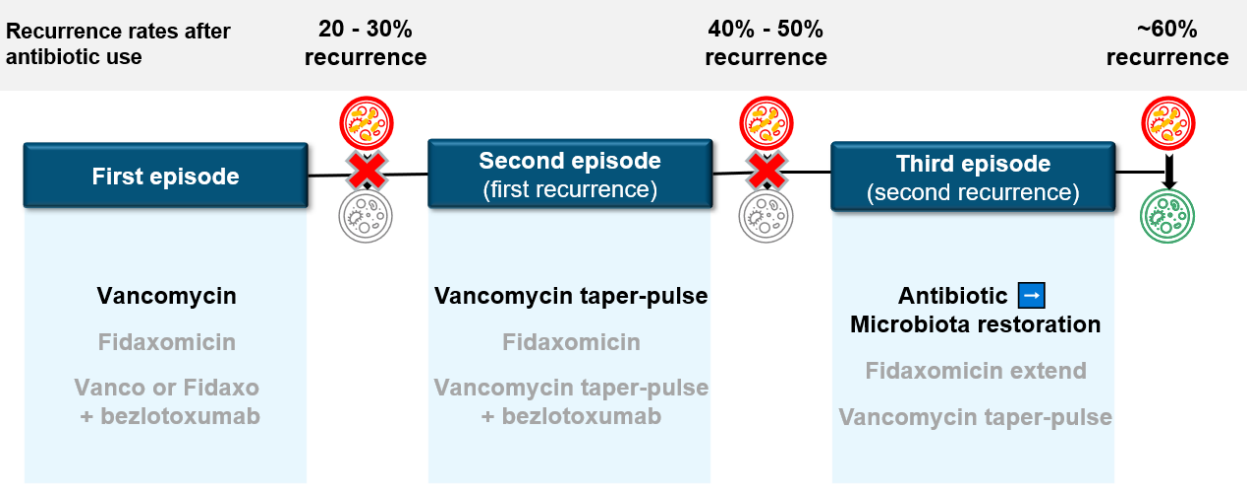
Several options are indicated to treat the first (or primary) episode of CDI in accordance with available treatment guidelines. These treatments include vancomycin or fidaxomicin, with vancomycin being the most prescribed option, despite its known disruption of the gut microbiome.

Fidaxomicin is relatively sparing of the gut microbiome and has lower recurrence rates compared to vancomycin, but it is not commonly used due to cost-benefit considerations. Bezlotoxumab is an FDA-approved treatment, which does not treat primary CDI, but it reduces the risk of recurrence when used in addition to standard of care antibiotics in patients who are at a high risk of recurrent disease.

Figure 3 depicts the current common treatment landscape for CDI. The standard of care for a first episode is antibiotic treatment, but up to 30% of patients will experience recurrent infection after the first episode. Second episode CDI are also commonly treated with an antibiotic, possibly taper-pulsed vancomycin, yet upwards of 50% of infections may still recur. For third and subsequent episodes, data consistently show a worsening cycle of recurrence after antibiotics, because the antibiotics can disrupt and do not restore the microbiome. Accordingly, recent updates to rCDI treatment guidelines include recommendations to consider FMT therapy, acknowledging its experimental and unapproved status. This underscores the significant patient need and emphasizes that microbiome-restorative approaches earlier in the recurrence progression are needed and desired by patients and physicians.

The rarity of rCDI makes it challenging to study patients with rCDI, but the seriousness of recurrent infection makes it imperative to find well-studied treatments.

Figure 3: Current Treatment Guidelines for CDI



(Johnson et al., 2021; C. R. Kelly et al., 2021a, 2021b)

2.2.1 Antibiotics

Antibiotic treatment is the current standard of care for treating CDI. However, antibiotic use has been linked to gastrointestinal dysbiosis, leading to negative clinical outcomes such as recurrence of CDI, and overuse has contributed to the rise of antibiotic-resistant pathogens (Guh et al., 2020; Khanna, 2021). The challenge of treating *C. difficile* is due to its fundamental biology, since it exists in both spore and vegetative forms. The vegetative form is the pathogen that proliferates

and produces Toxins A and B, which cause colitis and symptomatic infections. The spore form is a dormant but very resilient form of the pathogen. Because *C. difficile* spores are largely resistant to antibiotics, they can germinate into vegetative forms after antibiotic treatment has been discontinued.

These dynamics explain the fundamental challenge with CDI; treatment of an active infection with antibiotics does not eliminate the spore form of the organism, which may germinate into the vegetative form after completion of antibiotic therapy, causing a subsequent recurrence of the disease. According to clinical guidelines and the CDC, recurrent infections are distinguished from new infections as those that occur within 8 weeks of completing antibiotic therapy (C. R. Kelly et al., 2021a). Although most patients with a primary clinical episode of CDI respond to treatment with vancomycin or fidaxomicin, many patients experience several recurrent episodes. Once the disease has recurred, the risk of further recurrences is increased, with 40% to 60% of patients experiencing additional recurrent episodes (C. P. Kelly, 2012; Wenisch et al., 1996).

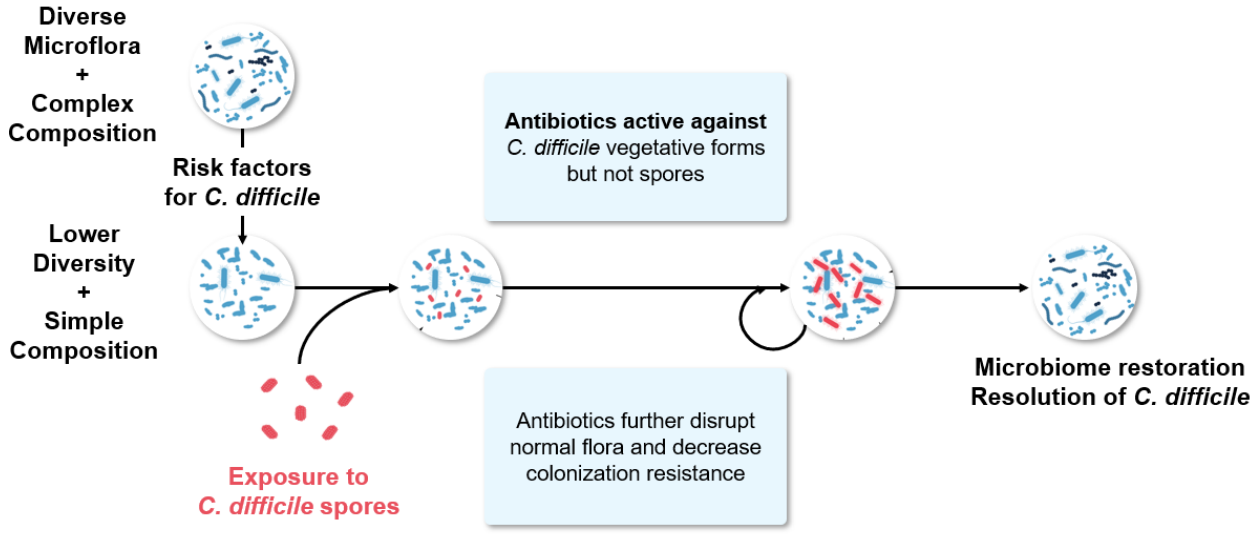
2.2.2 Monoclonal Antibodies

Bezlotoxumab, a monoclonal antibody against CDI Toxin B that must be administered as an intravenous infusion concurrent with antibiotic therapy, has demonstrated efficacy against future CDI recurrences. However, it carries the risk of heart failure, especially in patients with a history of congestive heart failure and should be reserved for when the benefits outweigh the risks (Merck & Co., 2016).

2.2.3 Importance of Restoring the Microbiome

Microbiome restoration is a viable approach to prevent *C. difficile* recurrence (Figure 4). Healthy patients have a diverse microbiome that can help resist colonization by pathogens like *C. difficile*. Upon exposure to risk factors for *C. difficile*, most prominently antibiotic therapy, the microbial diversity declines and the microbiome composition can change. In this state, *C. difficile* spores can germinate into vegetative forms, colonize, and produce the toxins that lead to diarrhea and other symptoms of CDI. The antibiotics that are used to treat CDI are active against the vegetative forms but not the spores. These antibiotics, especially vancomycin, are also active against the normal flora of the healthy microbiome (Tannock et al., 2010; Thorpe et al., 2018).

Figure 4: Restoring the Microbiome to Prevent CDI Recurrence



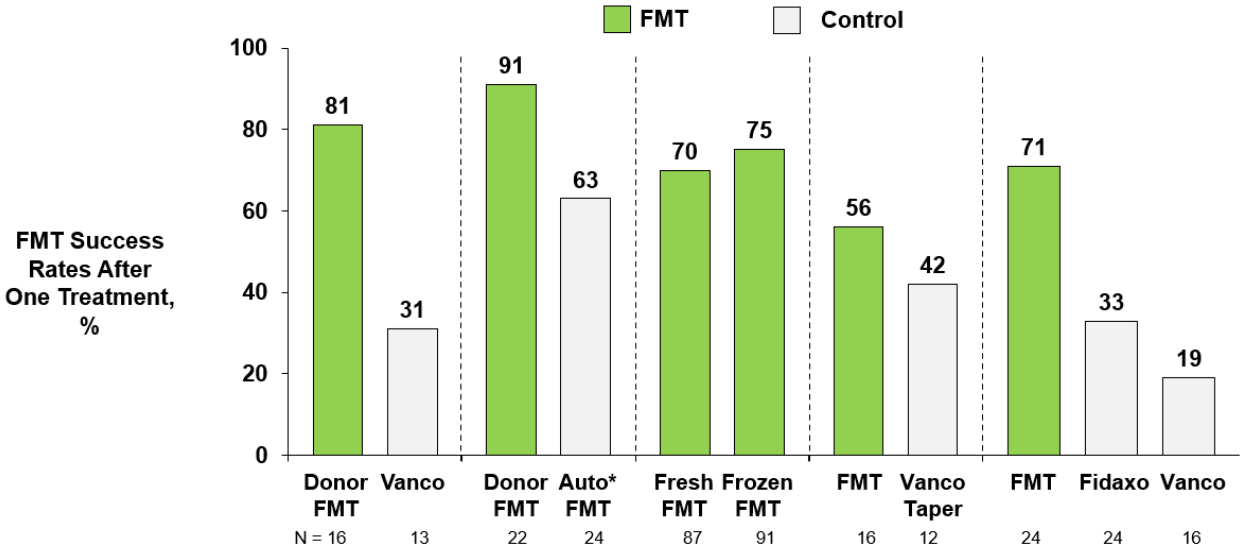
(Khanna, 2021)

2.2.4 Unapproved FMT

Despite using guideline-recommended antibiotic therapies, the rates of recurrence after a first episode are 20-30% and can be upwards of 60% after 3 or more infections. Restoration of the gut microbiome with a microbiome-based therapeutic often leads to resolution of CDI (Khanna, 2021). Therefore, FMT has become widely accepted as a treatment option for rCDI.

Although not being an FDA-approved therapy, the demand for FMT from patients and use by physicians is increasing. FMT has shown promising success rates among patients that have failed first- and second-line therapies, ranging from 56% to 91% in previous small, randomized controlled trials (Figure 5). This efficacy data supporting FMT has prompted updates to US treatment guidelines, which now recommend FMT for treatment and prevention of recurrent C. *difficile* infection after multiple recurrences (Johnson et al., 2021; C. R. Kelly et al., 2021). The concept of FMT is well established, with many sites around the US currently providing this treatment.

Figure 5: FMT Success Rates in Small Randomized Controlled Trials



*Autologous
(Hota et al., 2017; Hvas et al., 2019; C. R. Kelly et al., 2016; Lee et al., 2016; van Nood et al., 2013)

However, current FMT therapy has posed a number of safety challenges due to lack of standardization, related in part to its complexity and multiple components (Bafeta, Yavchitz, Riveros, Batista, & Ravaud, 2017). It remains dependent on human stool donations, but no FDA-mandated standards for donor screening or donation testing currently exist. Screening of donors should include general health and infection assessments to exclude those with symptomatic disease or known pathogens. Stool testing should include tests for enteric pathogens, viruses, parasites, and multi-drug resistant infections, such as extended-spectrum beta-lactamase (ESBL)-producing organisms. Additionally, donors should undergo blood tests for transmissible infections, including HIV, hepatitis, syphilis, and others. Any donor screening program should also be cognizant of emerging pathogens, such as the SARS-CoV-2 infection (Feuerstadt et al., 2021; Khanna, 2021; Khanna & Kraft, 2021; Khanna & Pardi, 2020). An approved product would allow for rigorous and consistent screening processes as well as safety surveillance to monitor the effectiveness of those processes.

In addition to safety concerns of unapproved FMT, the current lack of scalability limits the use of unapproved FMT for patients throughout the US. Also, the COVID-19 pandemic has diminished inventory of unapproved FMT, with most distribution restricting to emergency use only or dependent on individual physician development and administration, leaving many patients with rCDI without an available microbiome restoration therapy (Feuerstadt et al., 2021; Hota et al., 2017; Hvas et al., 2019; C. R. Kelly et al., 2016; Khanna, 2021; Khanna & Kraft, 2021; Khanna & Pardi, 2020; Lee et al., 2016; Tariq et al., 2017; van Nood et al., 2013).

2.2.5 Patient Unmet Medical Need

Patients with CDI may become involved in a cycle of recurrence because the antibiotics used to treat the disease may also be a risk factor for subsequent infection incurring increased morbidity, mortality, and healthcare expenditures. This group includes patients who fail to respond to standard therapies. Development of treatments aimed at restoring the microbiome for patients

with rCDI is urgently needed to fulfill this important unmet medical need. There would be clear benefits from the availability of an FDA-approved microbiome-based therapeutic. Physicians and patients want and need a well-studied, consistent product that has an established, positive benefit-risk profile. Approval of a standardized microbiome-based therapeutic would reduce variability and heterogeneity of the processes and preparation, improve access for this orphan patient population who suffer from a debilitating and potentially life-threatening condition; and finally give patients a means to actively address the cycle of recurrence.

3 PRODUCT DESCRIPTION

Summary

- RBX2660 is proposed as a treatment to reduce recurrence of CDI in adults following antibiotic treatment for recurrent *C. difficile* infections (rCDI).
- RBX2660 is a pre-packaged single dose 150 mL fecal microbiota suspension for rectal administration containing diverse spore-forming and non-spore-forming bacteria, including Bacteroides, and is biologically sourced, health screened, and pathogen tested to ensure patient safety.
- Standardized donor qualification and pathogen screening processes ensure product quality and patient safety.

3.1 Proposed Indication

The proposed indication for RBX2660 is to reduce the recurrence of CDI in adults following antibiotic treatment for rCDI.

3.2 Product Overview and Characteristics

3.2.1 Product Description

RBX2660 is a standardized, stabilized dosage form of a fecal microbiota suspension developed with the intent to reduce rCDI. RBX2660 is supplied as a pre-packaged, single-dose 150 mL fecal microbiota suspension containing a consortia of $1x10^8$ to $5x10^{10}$ CFU/mL of diverse viable bacteria, including Bacteroides. The final dosage form is delivered to patients via a single rectal administration of the liquid suspension that is instilled into the lower intestinal tract via an administration tube set (Figure 6).

Figure 6: RBX2660 Packaged Product

3.2.2 Quality System and Controls

As a drug product, compliance with regulatory requirements is provided throughout the product's lifecycle utilizing the required current Good Manufacturing Practices (cGMP), as well as quality controls specified under the Rebiotix Quality Management System. Given the nature of the product being a live biologic sourced from human donors, the product consistency is the result of the process consistency and related validated quality control test methods. The foundation of the Rebiotix QMS is structured to align with the requirements of FDA cGMPs for drugs (21 CFR parts 210 and 211) and biologics (21 CFR parts 600, 601 and 610), and incorporates the principles of a pharmaceutical quality system, as defined in ICH Q10 FDA (FDA, 2009, 2022a, 2022b, 2022c, 2022d, 2022e).

The donor screening, testing, manufacturing process, potency assays, and storage conditions established for the product have been developed with FDA oversight during the clinical program conducted under the Investigational New Drug (IND) application, and are under additional review by the FDA as part of the marketing application. These controls are appropriate for the proposed indication and patient population, and also align with the regulatory considerations published in the literature (Carlson, 2020) for products derived from human stool.

The QMS system includes detailed processes and procedures to ensure appropriate controls are in place for suppliers, materials, donor screening, DHS collection and testing, facilities, equipment, drug production and process controls, product testing and laboratory controls, packaging, labeling, storage and shipping conditions among others. Ongoing pharmacovigilance is also performed to provide continued safety monitoring and reporting. Together, these elements have provided a consistent drug product throughout the clinical program, from which to evaluate efficacy and safety.

3.2.3 Donor and Source Material Screening and Testing

Donor screening and DHS testing controls ensure that stool for drug product meets all of the DHS acceptance criteria and is obtained only from eligible, qualified donors.

Donors are recruited for long-term participation which provides a more consistent base of donors over time that must pass ongoing testing to remain in the program. Before being allowed to donate, donor candidates complete a health and lifestyle questionnaire and then provide blood, stool and nasopharyngeal swab samples for analysis for the presence of potential pathogens. Once all screening and testing is completed, and the results meet the acceptance criteria, the candidate is determined to be qualified for participation into the stool donation program. Once qualified, donors continually undergo routine screening for pathogens including ongoing blood, stool and SARS-CoV-2 testing. Each stool donation is tested for the presence of pathogens. In total, in addition to the Donor health questionnaires, before any drug product is eligible for release test results from blood tests both before and after the donation, multiple SARS-CoV-2 test results before and after the donation, and DHS test results for 29 different pathogens must all meet acceptance criteria. These tests are in addition to the quality controls for each lot of drug product establishing minimum criteria for potency, diversity and Bacteroides species growth.

The donor program has systems in place to monitor test results and emerging threats as part of the commitment to patient safety. Medical advisors for the program, who are physicians with relevant subject matter expertise, are required to provide clinical oversight of the screening program. These physicians review monitoring reports, risk assessments and any items related to donor testing or patient safety that require clinical input. The donor monitoring program also

includes surveying and collecting information from health agencies and news sources, including (but not limited to) the FDA, CDC, peer-reviewed publications and food safety sources. As part of our Quality Management System all processes, including the donor program, are monitored and updated to ensure patient safety and regulatory compliance and will continue to be updated as needed in the post-marketing setting. Planned post-marketing pharmacovigilance is described in Section 8.

3.2.4 Drug Product Manufacturing

After the donor and stool is screened, production of RBX2660 follows a detailed manufacturing and control strategy compliant with drug product cGMP requirements, as described above. One drug product batch is manufactured from one lot of DHS, defined as a single donation from a single donor. The number of doses in a drug product batch varies depending on the mass of the DHS, which can vary between lots. A high-mass lot of DHS will yield several doses while a low-mass lot may yield as little as one dose. This process enables us to maintain traceability from a single donor's donation to the individual lot and dose of drug product.

Potency of the dose of RBX2660 is measured using a validated assay to establish the product meets the acceptance criteria measured in CFU per mL. These methods have remained in place for the entirety of the clinical development program and have been validated for use for release of commercial product.

The drug substance is generated as a result of combining DHS and a solution of polyethylene glycol 3350 (PEG) and 0.9% sodium chloride irrigation (see Section 3.2.5 for additional details on excipients). The drug product is the drug substance filled into the primary container (ethylene vinyl acetate bag) containing 150 mL to 170 mL of drug substance.

RBX2660 is not a sterile product given it consists of living bacterial organisms and no antimicrobial preservatives are utilized in the formulation. There is a comprehensive contamination and control strategy in place under the quality system to ensure that all handling and manufacturing of the product is done in a highly controlled environment in a biological safety cabinet with effective training, cleaning and monitoring in place. These controls are audited, consistent with cGMP requirements and are in place so the product is not affected by the manufacturing environment, processes or storage.

RBX2660 is stored in ultra-cold conditions (-60 to -90°C) after production. The product is maintained at ultra-cold conditions through the distribution process and is transported using ultra-cold validated shipping conditions. Upon receipt at the prescriber's facility the product can be removed from the shipper, thawed at refrigerated conditions, and administered to the patient.

3.2.5 Nonclinical Information

The FDA agreed that non-clinical studies were not required for RBX2660. There are no nonclinical guidelines regarding the approval of human fecal microbiota as a medicinal product. Hence, a case-by-case approach is warranted when assessing the nonclinical safety of such products. Human stool transplantation is not considered to be absorbed into the body, and therefore standard pharmacokinetic and toxicology studies according to ICH S6 were not required.

The excipients used in the drug product are saline and polyethylene glycol 3350 (PEG). Polyethylene glycols are widely used in gastroenterology, as topical formulation excipients, and are listed on FDA's Inactive Ingredient Search for Approved Drug Products. PEGs display an inverse relation between molecular mass and intestinal absorbability, with practically no intestinal

absorption and minimal metabolism at molecular masses exceeding 3000. High molecular weight PEGs such as PEG 3500 display a benign toxicity profile, with no irritant potential. Hence GI local tolerance testing of the drug product is considered unnecessary.

4 REGULATORY AND DEVELOPMENT HISTORY

Summary

- The clinical development program for RBX2660 consisted of 6 studies, including 2 placebo-controlled studies (the Phase 2B Study 2014-01 and Pivotal Phase 3 Study 2017-01), 3 open-label studies (Phase 2 Studies 2013-001 and 2015-01, and the ongoing Phase 3 Study 2019-01), and 1 retrospective study (Study 2019-02).
- The results of the Phase 3 Study 2017-01 constitute the primary evidence of efficacy.
- The Phase 3 2017-01 study was designed to prospectively include data from the Phase 2B Study 2014-01, also a placebo-controlled, randomized study, through a Bayesian hierarchical model.
- The Phase 2B 2014-01 study supported the selection of 1 dose of RBX2660 as the regimen used in Study 2017-01.

4.1 Regulatory Milestones

Key regulatory milestones in the development of RBX2660 include:

- Fast Track designation granted in 2013.
- Orphan Drug designation received in 2014.
- Breakthrough Therapy designation granted in 2015.

4.2 Clinical Development Program

The clinical development program for RBX2660 consisted of 6 studies, including 2 placebo-controlled studies (the Phase 2B Study 2014-01 and Pivotal Phase 3 Study 2017-01), 3 open-label studies (Phase 2 Studies 2013-001 and 2015-01, and the ongoing Phase 3 Study 2019-01), and 1 retrospective study (Study 2019-02). All of these studies, placebo-controlled and uncontrolled, contribute to the overall totality of evidence supporting the safety and efficacy of RBX2660 (Figure 7).

Originally, 2 separate Phase 3 studies with a target enrollment of 300 patients each were planned. It was expected that at least 40 clinical study sites would be necessary for each study, due to the low prevalence of this disease with an orphan designation. However, the increased availability of FMT products made it difficult to enroll patients into the placebo-controlled Study 2017-01, even after substantial increases in the number of study sites.

While Study 2017-01 eventually achieved the number of planned patients, the slower than expected enrollment strongly suggested that an additional Phase 3 study would take at least 6 additional years to complete and would not be feasible in the current treatment environment for this serious and rare disease. The FDA acknowledged these extenuating circumstances, and given the orphan indication, the Agency proposed exploration of other approaches such as a Bayesian statistical design, thus allowing for a single Phase 3 trial as the basis of approval. As a result, a new statistical analysis plan incorporating a Bayesian design was incorporated in agreement with FDA. This approach is described in more detail in Section 6.4.1.5.2. The final number of participants in the clinical program also exceeded the FDA's required number of patients exposed to RBX2660 for safety assessment (N=600 target minimum; actual number 978); robust for orphan designated population.

The results of the randomized, blinded, placebo-controlled Phase 3 Study 2017-01 constitute the primary evidence of efficacy. The Phase 2B Study 2014-01, also a randomized, placebo-controlled study, was formally integrated into Study 2017-01 through a Bayesian hierarchical model, and also supported the selection of 1 dose of RBX2660 as the regimen used in Study 2017--01.

The nonrandomized, open-label studies (Studies 2013-001, 2015-01, and 2019-01), and the retrospective Study 2019-02 provide supplementary evidence of efficacy and safety for RBX2660. Studies 2019-01 and 2019-02 enrolled a broader patient population, including some patients with IBS, IBD, and immunocompromised conditions, who may not have qualified for enrollment, given the stricter eligibility criteria, in other studies.

Key design features of the randomized, placebo-controlled studies (Studies 2014-01 and 2017-01) and of the nonrandomized, open-label studies (Studies 2013-001, 2015-01, 2019-01 and 2019-02) are presented in Table 1 and Table 2, respectively.

All studies were conducted in the US and Canada in adults ≥18 years of age with documented rCDI, and the prospective studies required patients to be on standard of care oral antibiotic therapy prior to initial treatment with RBX2660.

In all studies except 2015-01, a second course of treatment (ie, open-label RBX2660) was allowed if the patient experienced a CDI recurrence after the first course of treatment. Antibiotic therapy was not administered prior to a second course of treatment in study 2013-001, but antibiotics were optional prior to a second course of treatment in studies 2014-01, 2017-01, and 2019-01.

Figure 7: RBX2660 Clinical Development Program

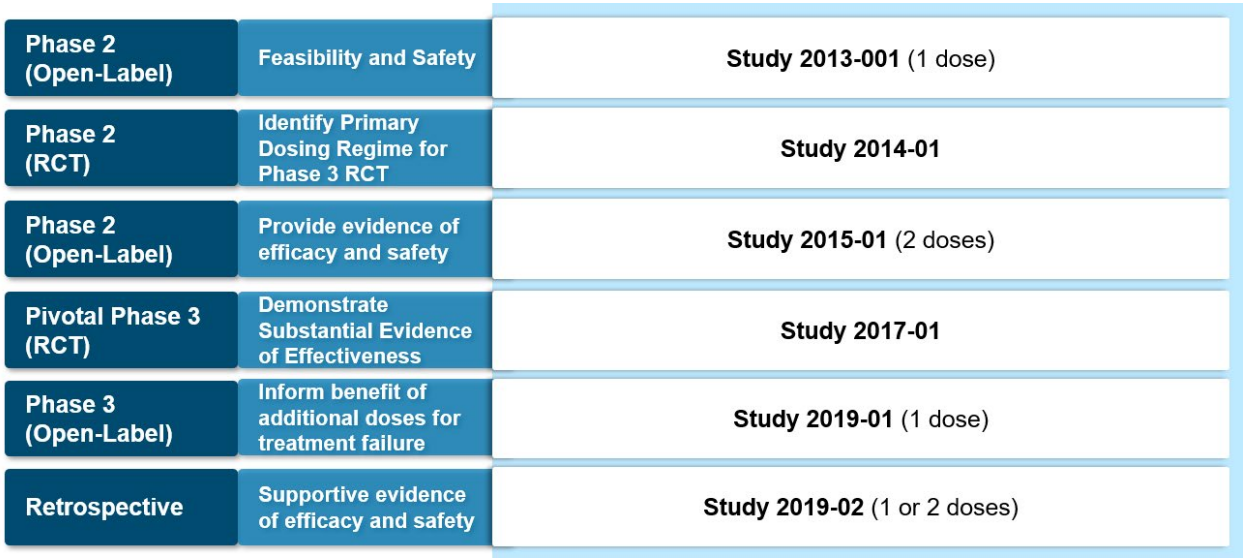


Table 1: RBX2660 Clinical Development - Key Study Design Features by Randomized, Placebo-Controlled Study

Randomized, Placebo-Controlled Studies		
Study Design Feature	**Study 2014-01 (Phase 2b)**	**Study 2017-01 (Phase 3)**
Total enrolled	150	320
Total randomized	133	289
Study design	Prospective, multicenter, double-blind, randomized, placebo-controlled, dose-regimen evaluation	Prospective, multicenter, double-blind, randomized, placebo-controlled
Primary endpoint	Treatment Success – Absence of CDI diarrhea for 8 weeks after study treatment (ITT primary efficacy population)	Treatment Success – Absence of CDI diarrhea for 8 weeks after study treatment (mITT primary efficacy population)
Comorbidities commonly associated with rCDI allowed[a]	None	None
Number of previous CDIs, including primary episode	Either: a) ≥2 recurrences of CDI after a primary episode and has completed ≥2 rounds of SOC oral antibiotic therapy; or b) has had at least 2 episodes of severe CDI resulting in hospitalization.	Either: a) ≥1 recurrence of CDI after a primary episode and had completed ≥1 round of SOC oral antibiotic therapy; or b) had at least 2 episodes of severe CDI resulting in hospitalization within the last year.
Efficacy Endpoint Adjudication	DSMB (blinded when adjudicating; Provided independent confirmation of success/failure for study reporting purposes)	EAC (provided independent blinded adjudications of Treatment Success or failure that were used for study analysis and reporting)
Antibiotic washout	24 to 48 hours	24 to 72 hours
Randomization and treatment groups/ treatment dose/treatment regimen	1:1:1 ratio RBX2660 2 Doses Group Placebo Group (2 doses placebo) RBX2660 1 Dose Group (1 dose RBX2660/1 dose placebo) 2 enemas administered 7 ± 2 days apart	2:1 ratio: RBX2660 (1 dose) Placebo (1 dose) 1 enema administered
Optional second treatment course?	Yes; up to 2 doses per treatment course	Yes; 1 dose per treatment course
Stool test for recurrence	Local laboratory	Central laboratory
Follow-up duration (months)	24[b]	6
Key contributions to development program	Regimen-finding Supplemental data for efficacy and 24 months safety follow-up.	Primary evidence of efficacy and persistence of efficacy; safety

CDI = *Clostridioides difficile* infection; DSMB = Data Safety Monitoring Board; EAC = Endpoint Adjudication Committee; IBD = inflammatory bowel disease; IBS = irritable bowel syndrome; SOC = standard of care.

[a] Includes IBD (ulcerative colitis, Crohn's disease), IBS, microscopic colitis, celiac disease, and immunocompromised conditions.

[b] Efficacy outcomes were only evaluated up to 8 weeks after the last enema; follow-up was performed past the efficacy endpoint of 8 weeks to collect data for safety evaluations.

Table 2: RBX2660 Clinical Development - Key Study Design Features by Nonrandomized, Open-Label Study

Study Design Feature	Nonrandomized, Open-Label Studies			
	2013-001 (Phase 2)	**2015-01 (Phase 2)**	**2019-01 (Phase 3)**	**2019-02 (Retrospective)**
Total enrolled	40	162	551	94
Study design	Prospective, multi-center, open-label, nonrandomized	Prospective, multi-center, open-label, historical controls	Prospective, multi-center, open-label, nonrandomized	Retrospective, multi-center, open-label, nonrandomized
Common comorbidities allowed[a]	None	None	Yes	Yes
Number of previous CDIs, including primary episode	Either a) ≥2 recurrences of CDI after a primary episode and has completed ≥2 rounds of SOC oral antibiotic therapy; or b) has had at least 2 episodes of severe CDAD resulting in hospitalization	Either a) ≥2 recurrences of CDI after a primary episode and has completed ≥2 rounds of SOC oral antibiotic therapy; or b) has had at least 2 episodes of severe CDI resulting in hospitalization	Investigator discretion, patients were required to have current diagnosis of rCDI or 2 episodes of severe CDI leading to hospitalization	Investigator discretion
Antibiotic washout	24 to 48 hours	24 to 48 hours	24 to 72 hours	Not applicable
Efficacy Endpoint Adjudication	None (investigator assessment only)	None (investigator assessment only)	EAC	None (investigator assessment only)
Treatment dose/treatment regimen	RBX2660 (1 dose; option of second dose)	RBX2660 (2 doses) administered 7 ± 2 days apart	RBX2660 (1 dose, with option of second dose if there was Treatment Failure)	RBX2660 (1 or 2 doses)
Optional second treatment course?	Yes	No	Yes	Investigator discretion
Stool test for recurrence	Local laboratory	Local laboratory	Central laboratory	Local laboratory
Follow-up duration (months)	6[b]	24[b]	6	6
Key contributions to development program	Feasibility; safety	Supportive evidence of efficacy; safety	Supportive evidence of efficacy and persistence of efficacy; expanded rCDI patient population (eg, IBD, IBS, and Immuno-compromised); safety	Supportive evidence for efficacy and persistence of efficacy; expanded rCDI patient population; safety

CDI = *Clostridioides difficile* infection; DSMB = Data Safety Monitoring Board; EAC = Endpoint Adjudication Committee; IBD=inflammatory bowel disease; IBS=irritable bowel syndrome; rCDI=recurrent *Clostridioides difficile* infection; SOC = standard of care.

[a] Includes IBD (ulcerative colitis, Crohn's disease), IBS microscopic colitis, celiac disease, and immunocompromised conditions.

[b] Efficacy outcomes were only evaluated up to 8 weeks after the last enema.

5 CLINICAL PHARMACOLOGY

RBX2660 is for rectal use only. Because the fecal microbiota suspension is not systemically absorbed, standard pharmacokinetic and toxicology studies are not warranted. However, microbiome analyses (eg, changes in microbial composition, concentration of vancomycin-resistant *Enterococcus*, and presence of *C. difficile*) were explored ad hoc in Phase 2 studies and were prospectively defined as exploratory endpoints in Phase 3 Studies 2017-01 and 2019-01.

The mechanism of action of RBX2660 is thought to involve repopulation and restoration of the composition and diversity of the gut microbiome to suppress *C. difficile* colonization and thus prevent CDI recurrence. In Studies 2017-01 and 2019-01, Treatment Success was associated with a shift of the gut microbiome from before to after RBX2660 administration. Before treatment patient microbiomes were dysbiotic—characterized by decreased diversity, decreased relative abundance of Clostridia- and Bacteroidia-class bacteria, and increased Gammaproteobacteria and Bacilli-class bacteria relative to most published healthy populations. Success was associated with a shift to compositions with increased diversity, increased relative abundance of Clostridia- and Bacteroidia-class bacteria, and decreased Gammaproteobacteria- and Bacilli-class bacteria.

6 CLINICAL EFFICACY

Summary

- Study 2014-01 was a Phase 2B, prospective, multicenter, randomized, double-blind, placebo-controlled study, evaluating the efficacy and safety of different dose regimens of RBX2660 in adults with rCDI.
  - Patients were randomized 1:1:1 to 2 doses of RBX2660, 1 dose of RBX2660 followed by placebo, and 2 doses of placebo; the doses in each group were given 1 week apart. A total of 133 patients were randomized (2-Dose RBX2660 Group=45 patients; 1-Dose RBX2660 Group=44 patients; and Placebo Group=44 patients).
  - The primary efficacy endpoint was Treatment Success, defined as the absence of CDI diarrhea for 8 weeks after study treatment (ITT primary efficacy population).
  - The primary comparison of the 2-Dose RBX2660 Group (N=25/45; 55.6% Treatment Success) to the Placebo Group (N=19/44; 43.2% Treatment Success) in the blinded portion of the study in the ITT population was not significant (p=0.243).
  - No meaningful difference was observed in 1 dose of RBX2660 (Treatment Success, 56.8% [25/44]) and 2 doses of RBX2660 administered 1 week apart (Treatment Success, 55.6% [25/45]); therefore, a single dose of RBX2660 was selected for the Pivotal Phase 3 Study.
- Pivotal Study 2017-01 was a prospective, multicenter, randomized, double-blinded, placebo-controlled Phase 3 study to confirm the efficacy and evaluate the safety of RBX2660 against placebo for the prevention of rCDI.
  - Randomization was at a 2:1 ratio to 1 dose of either RBX2660 or placebo; a total of 193 patients were randomized to RBX2660 treatment and 96 patients randomized to placebo.
  - The primary efficacy endpoint was the absence of CDI diarrhea for 8 weeks after study treatment in the mITT analysis set.
  - The study was analyzed using a Bayesian hierarchical model, i.e., incorporating dynamic borrowing of data from the Phase 2B Study 2014-01.
  - The planned primary Bayesian analysis resulted in a model-estimated Treatment Success rate of 70.4% in the RBX2660 group and 58.1% in the Placebo group in the mITT population, with an estimated treatment difference of 12.3 percentage points (95% credible interval [CrI]: 1.4 to 23.3) and a probability of superiority to placebo of 98.6%.
  - At FDA's request during BLA review, the integrated Bayesian analysis of the primary efficacy endpoint analysis aligned the analysis populations and definitions between Studies 2014-01 and 2017-01.
    - The model-estimated Treatment Success rate was 70.6% in the RBX2660 group and 57.5% in the Placebo group in the mITT population, with an estimated treatment difference of 13.1 percentage points (95% CrI: 2.3 to 24.0) and a posterior probability of superiority of 99.1%.
  - Results of Treatment Success in subgroups were generally consistent with the primary analysis, including analyses by age, sex, and race.
  - Among the eligible patients who had been treated with RBX2660 during the blinded period of the study but had Treatment Failure, 41 patients elected a second course of RBX2660, and more than half (22/41; 53.7%) of those patients reported Treatment Success after the additional 8 weeks.

6.1 Overview of Study Designs

All studies were conducted at multiple sites in the US and Canada. Patients were adults (≥ 18 years) and typically enrolled into the prospective studies while taking SOC oral antibiotic therapy. The clinical study populations were reflective of common clinical practice, as documented in

published recommended clinical treatment guidelines (Johnson et al., 2021; C. R. Kelly et al., 2021a). Consistent efficacy was shown across the studies.

RBX2660 treatment was completed following an antibiotic washout period of 24 to 72 hours. Depending on the study, the first course of treatment consisted of 1 or 2 doses of RBX2660 (or placebo), followed by monitoring for recurrence of CDI within 8 weeks after last dose. Treatment Success was defined as the absence of CDI symptoms, primarily CDI diarrhea, and no need for re-treatment within 8 weeks after administration of the last assigned study dose. The 8-week timepoint is a well-accepted differentiator of recurrences from new infections (ie, new occurrences) (C. R. Kelly et al., 2021a).

Based on the 8-week timepoint, patients who had a confirmed recurrence of CDI within 8 weeks were considered Treatment Failures and, in all studies except for 2015-01, were offered an open-label treatment course of RBX2660. Regardless of treatment outcome, patients were monitored through 6 or 24 months after receiving the last dose of RBX2660, as defined in the study protocols.

6.2 Phase 2 Study 2013-001 (Open-Label, First-in-Human Study)

The Phase 2 Study 2013-001 was an open-label, first-in-human trial, conducted in 34 treated patients who received 1 dose of RBX2660, with the option for a second treatment course after CDI recurrence. Patients were followed for 6 months to evaluate safety. The primary efficacy endpoint was Treatment Success, defined as the absence of CDAD (passage of 3 or more unformed stools in ≤ 24 hours for at least 2 consecutive days) at 8 weeks after receipt of RBX2660.

The efficacy analysis for patients in the group with Available Data (treated patients with non-missing data for each specific endpoint, i.e., either the patient experienced an event or completed the associated follow-up without the occurrence of an event) are presented in Table 3. Sixteen of 32 patients (50.0%) were considered a Treatment Success after their first treatment with RBX2660. Overall Treatment Success was 87.1% (27/31) after a second treatment course of RBX2660.

Based on these data, the RBX2660 treatment regimen was next assessed to determine if a 1-dose regimen or 2-dose regimen, administered 1 week apart, would be optimal for the treatment of rCDI.

Table 3: Phase 2 Study 2013-001 – Patients Considered a Treatment Success at 8 Weeks after Receiving Last Dose of RBX2660 – Available Data

Treatment Success	RBX2660
Overall Treatment Success at 8 weeks after last dose of RBX2660*	
n/N (%)	27/31 (87.1)
95% CI	(70.2, 96.4)
Patients considered Treatment Success at 8 weeks after first dose of RBX2660	
n/N (%)	16/32 (50.0)
95% CI	(31.9, 68.1)
Patients considered Treatment Success at 8 weeks after second dose of RBX2660	
n/N (%)	11/14 (78.6)
95% CI	(49.2, 95.3)

*Note: Of the 16 patients who failed their first treatment, 15 patients proceeded to receive a second enema of RBX2660. Of those, one patient was not counted because she did not reach the 8 week endpoint assessment and therefore results are presented for 31 patients.

6.3 Phase 2B Study 2014-01

6.3.1 Investigational Plan

6.3.1.1 Overall Design

Study 2014-01 was a prospective, multicenter, randomized, double-blinded, placebo-controlled, 3-arm Phase 2B study evaluating the efficacy and safety of RBX2660 for the prevention of rCDI. This study was used to determine the treatment regimen of RBX2660 for subsequent confirmation in a pivotal Phase 3 trial. A schematic of the study design is shown in Figure 1.

Up to 150 patients were to be randomized and treated in the study. Randomization was 1:1:1 to 2 doses of RBX2660, 1 dose of RBX2660 followed by 1 dose of placebo, and 2 doses of placebo. In each treatment group, doses were administered 7 ± 2 days apart.

Patients who were deemed Treatment Failures following the blinded treatment course, per the prespecified Treatment Failure definition, could choose to receive a second treatment course with up to 2 open-label RBX2660 doses administered 7 ± 2 days apart.

A primary efficacy analysis and safety analyses were conducted when all randomized patients had completed their 8-week follow-up assessment after receiving the blinded treatment. The final reporting was done when all patients had completed the 24 months follow-up.

6.3.1.1.1 Definitions of Treatment Success and Treatment Failure

Definitions

Treatment Success was defined as:

- The absence of CDI diarrhea (passage of 3 or more unformed stools in 24 or fewer consecutive hours for at least 2 consecutive days) for 8 weeks after completing a study treatment.

Treatment Failure (CDI recurrence) was defined as:

1) The presence of CDI diarrhea, with or without other CDI symptoms, at < 8 weeks after administration of the last assigned study dose;
2) A positive stool test for *C. difficile*;
3) Need for re-treatment for CDI; and
4) No other cause for CDI symptoms had been determined.

Determination of Success/Failure (Investigator and DSMB)

The site investigator made the initial determination of success or failure based on the predefined study definitions. The site investigator's assessment was then provided to the independent Data and Safety Monitoring Board (DSMB) for blinded adjudication of each patient's treatment outcome status (Treatment Success, Treatment Failure, or Indeterminate), which was utilized for study analysis and reporting purposes. Patients who met some but not all four criteria for Treatment Failure were included under the category Indeterminate and counted as Treatment Failures for purposes of efficacy analysis.

Management of Treatment Failures

Following blinded treatment, patients with Treatment Failure could elect to be scheduled for administration of up to 2 open-label RBX2660 doses (administered 7 ± 2 days apart) within 56 days of completion of blinded study treatment. The use of antibiotics prior to open-label RBX2660 treatment was at the discretion of the investigator.

If a patient received open-label RBX2660, the follow-up requirements restarted from the day of administration of the last open-label RBX2660 dose according to the same schedule as required for the blinded portion of the study.

Treatment Failures who did not receive an open-label RBX2660 dose were to continue to follow their original schedule of assessments for the duration of the study based on the last blinded study dose.

6.3.1.2 Randomization, Treatments, and Study Dosing Regimen

For the blinded treatment, the randomization schedule was created using randomized blocks within 3 strata, based on antibiotics used at screening (Vancomycin, Fidaxomicin, or Other). Randomized patients were assigned to one of 3 dosing regimens consisting of 2 doses administered 7 ± 2 days apart:

- 2 doses of RBX2660,
- 1 dose of RBX2660 followed by 1 dose of placebo; and
- 2 doses of placebo.

Active treatment was RBX2660 (fecal microbiota suspension) in a dose of 150 mL supplied in an enema bag.

The placebo dose consisted of 150 mL of normal saline and cryoprotectant in the same formulation as RBX2660 in an enema bag, but without the fecal microbiota suspension.

Duration of Treatment

Patients were expected to participate in the study for approximately 25 months: 1 month enrollment and 24 months follow-up.

6.3.1.3 Objectives and Endpoints

6.3.1.3.1 Study Objectives

The primary study objective was to assess the efficacy of 2 doses of RBX2660 vs. 2 doses of placebo.

Secondary objectives included:

- To evaluate the efficacy of (1 dose of RBX2660 and 1 dose of placebo) vs. 2 doses of placebo.
- To evaluate the efficacy of 2 doses of RBX2660 vs. (1 dose of RBX2660 and 1 dose of placebo).

6.3.1.3.2 Primary Endpoint

The primary endpoint of Treatment Success was defined as the absence of CDI diarrhea for 8 weeks after study treatment in the 2-Dose RBX2660 group vs. Placebo group (2 doses placebo).

6.3.1.4 Selection of Study Population

Inclusion criteria:

1. ≥ 18 years old.
2. Medical record documentation of rCDI either: a) at least 2 recurrences after a primary episode and had completed at least 2 rounds of standard of care oral antibiotic therapy or b) had at least 2 episodes of severe CDI resulting in hospitalization.
3. Already taking or was starting a course of antibiotics to control rCDI symptoms at the time of enrollment. [Note: Patient's rCDI symptoms must have been controlled (<3 loose stools/day) while taking this course of antibiotics].
4. A positive stool test for the presence of *C. difficile* within 60 days prior to enrollment.
5. Willing and able to have an enema(s).
6. Completed the stool and serum testing required for the study.
7. Agreed to abstain from non-dietary probiotics for the duration of the study.
8. Agreed to abstain from vancomycin, metronidazole, fidaxomicin, rifaximin, nitazoxanide and intravenous immune globulin for the duration of the study unless prescribed to treat rCDI.
9. Agreed to practice a form of effective contraception during study participation.
10. Had a negative urine pregnancy test at the time of enrollment and on the day of each enema administration (females of child-bearing potential only).
11. Willing and able to provide informed consent and Health Insurance Portability and Accountability Act (HIPAA) authorization.
12. Willing and able to complete the required Patient Diary.
13. Willing and able to meet all study requirements, including attending all assessment visits and phone calls.

Exclusion criteria:

1. A known history of continued *C. difficile*-associated diarrhea despite being on a course of antibiotics prescribed for CDI treatment.
2. Required antibiotic therapy for a condition other than CDI.
3. Previous FMT prior to study enrollment.
4. History of IBD, eg, ulcerative colitis, Crohn's disease, or microscopic colitis.
5. Diagnosis of IBS as determined by Rome III criteria.
6. History of chronic diarrhea.
7. History of celiac disease.

8. Disease symptoms caused by a confirmed intestinal pathogen other than *C. difficile*.
9. Colostomy.
10. Intraabdominal surgery within the last 60 days.
11. Evidence of active, severe colitis.
12. History of short gut syndrome or motility disorders.
13. Requires the regular use of medications to manage bowel hypermotility.
14. Planned therapy in the next 3 months that may cause diarrhea (eg, chemotherapy).
15. Planned surgery requiring perioperative antibiotics within 6 months of study enrollment.
16. Life expectancy of < 12 months.
17. Compromised immune system (eg, HIV infection; AIDS-defining diagnosis or CD4 <200/mm^3; inherited/primary immune disorders; immunodeficient or immunosuppressed due to a medical condition or medication; current or recent (< 90 days) treatment with chemotherapy; or current or recent (< 90 days) treatment with immunosuppressant medications).
18. Taking systemic steroids (≥ 20 mg a day or prednisone-equivalent) or is expected to be on steroids after enrollment through 8 weeks after completing the assigned study treatment.
19. An absolute neutrophil count of < 1000 cells/µL.
20. Known or suspected current (< 90 days) illicit drug use.
21. Pregnant, breastfeeding, or intended to become pregnant during study participation.
22. Participating in a clinical trial of another investigational product (drug, device or other) and had not completed the required follow-up period.
23. Patient, in the opinion of the investigator, for whatever reason, should be excluded from the study.

6.3.1.5 Statistical and Analytic Plans

6.3.1.5.1 Data Sets Analyzed

There were 4 analysis populations:

- **Safety Population (SP):** The SP was defined as the population of randomized patients who received any study treatment. Patients were analyzed according to the treatment they actually received.
- **Intent-to-Treat (ITT):** The ITT population consisted of all randomized patients, regardless of whether they completed their assigned study treatment. Patients were analyzed according to the randomized treatment rather than the actual treatment received should any treatment misallocations or discontinuations occur.
- **Modified Intent-to-Treat (mITT):** The mITT population was defined as the ITT population who completed at least one dose of study treatment, regardless of treatment received, but excluding patients who discontinued from the study during the blinded period, prior to

evaluation of Treatment Failure or Success, for any reason and excluding patients who had deviations from any inclusion or exclusion criteria.

- **Per Protocol (PP):** The PP population consisted of all ITT patients who received the treatment to which they were randomized and were evaluable for Treatment Success at 8 weeks after the last assigned treatment, excluding patients who withdrew consent or were lost to follow-up during the double-blind period, prior to evaluation of Treatment Failure or Success; patients who expelled a moderate or large amount of dose; patients who were adjudicated by the DSMB as indeterminate; and patients who had major protocol deviations as determined by a clinical review of patient data prior to database lock; and patients that had eligibility criteria deviations.

6.3.1.5.2 Efficacy Analysis

The primary efficacy analysis was prespecified in the Statistical Analysis plan to be performed on the ITT analysis set. The primary comparison was between the 2-Dose RBX2660 group and the Placebo group with other comparisons possible using a closed hierarchical testing method. The two-sided alpha level for the primary efficacy analysis was 0.05. If the null hypothesis that the success rates for these 2 groups are equal was rejected, the 1-Dose RBX2660 group would be compared to the Placebo group using a two-sided alpha level of 0.05. The next step in the hierarchical testing was to compare the 2-Dose RBX2660 group vs. the 1-Dose RBX2660 group. Treatment groups were compared using Pearson's chi-square test and the treatment difference presented with a 95% confidence interval based on the normal approximation.

6.3.1.5.3 Handling of Missing Data for the Primary Endpoint

Randomized patients who did not complete the assigned study treatment were considered Treatment Failures. Patients who discontinued the study prior to 8 weeks after administration of the last assigned study dose during the blinded period for any reason were considered Treatment Failures.

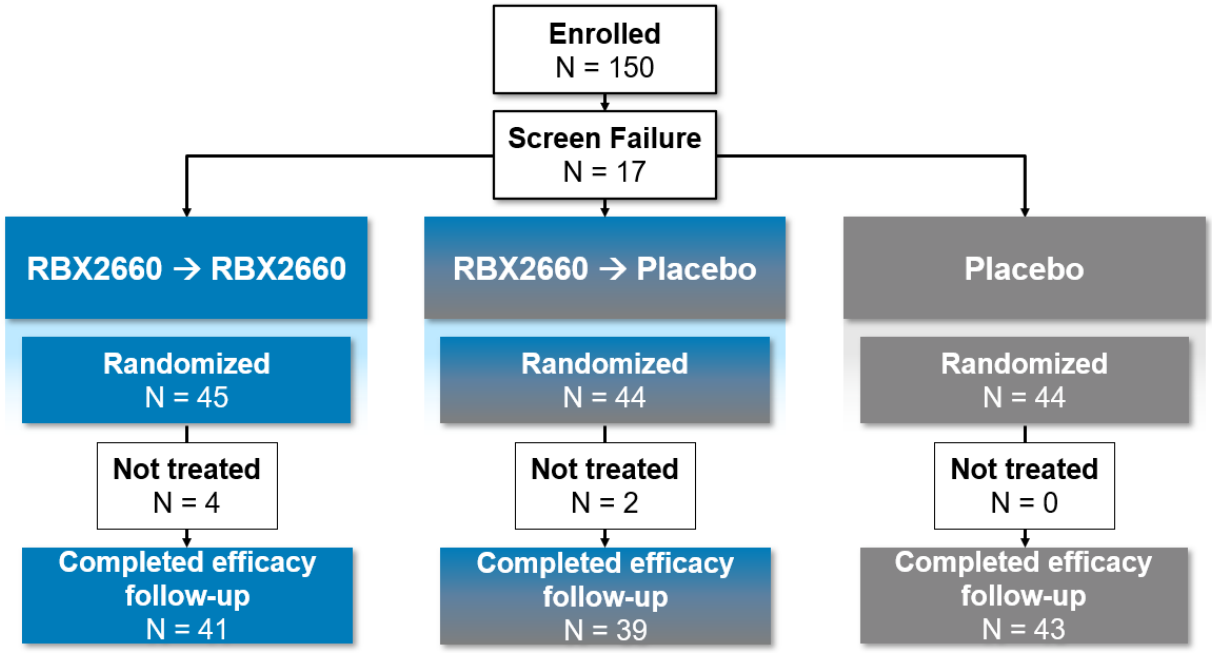
6.3.2 Study Patients

6.3.2.1 Disposition

A total of 150 patients were enrolled in Study 2014-01 across 21 clinical sites in the US and Canada (Figure 8). A total of 17 enrolled patients were screen failures, did not proceed to randomization and were removed from the study. Of the 133 randomized patients, 5 patients were withdrawn prior to treatment for a variety of reasons, including withdrawal by patient or investigator, and 1 patient who died prior to receiving study treatment. Among the 5 patients withdrawn prior to treatment, 1 was later re-enrolled, randomized and treated. In total 128 randomized patients were exposed to blinded dose, with 127 patients successfully completing at least one blinded dose; one patient did not receive the full dose due to anxiety, and elected to withdraw from the study.

After receiving one blinded dose, 14 patients withdrew for various reasons. One remaining patient was documented as missing the second dose due to protocol deviation, and subsequently continued follow-up without receiving a second blinded dose. In total, 123 patients completed the efficacy follow-up.

Figure 8: Phase 2B Study 2014-01 – Patient Disposition



RBX2660/RBX2660 = 2 doses, RBX2660/Placebo = 1 dose
Note: Regarding the 2 patients classified as "not treated" in the RBX2660/Placebo group, 1 did not complete treatment, but is included in the safety population because of partial exposure to study treatment.

6.3.2.2 Baseline Demographics and Characteristics

There were no notable differences in the baseline demographics among the treatment arms (Table 4). Baseline demographic characteristics were generally representative of an adult patient population with rCDI and were consistent across treatment groups. The mean age ranged from 59 to 64 years, the majority of patients were female, and were mostly White.

Table 4: Phase 2B Study 2014-01 - Baseline Demographics and Characteristics (ITT Analysis Set)

	2-Dose RBX2660 (RBX-RBX) N = 45	1-Dose RBX2660 (RBX-PBO) N = 44	Placebo (PBO-PBO) N = 44
Age (years), mean (SD) Min, max	63.6 (19.2) (24 – 89)	61.0 (19.7) (18 – 88)	58.8 (19.2) (19 – 92)
Female, n (%)	26 (57.8%)	25 (56.8%)	30 (68.2%)
White, n (%)	44 (97.8%)	42 (95.5%)	43 (97.7%)
Duration of CDI (days), mean (SD)	18.8 (13.4)	17.3 (11.4)	19.8 (17.7)
Previous episodes of CDI*, mean	4.3	4.1	3.8
Hospitalization			
Due to CDI episode, n (%)	26 (57.8%)	19 (43.2%)	25 (56.8%)
Duration (days), median (IQR)	9.5 (15.0)	7.0 (6.0)	5.0 (3.5)
Vancomycin during screening, n (%)	41 (91.1%)	38 (86.4%)	40 (90.9%)

*Inclusive of qualifying CDI event

6.3.3 Results: Primary and Secondary Efficacy Endpoints

In this Phase 2B regimen-finding study, the primary endpoint comparing Treatment Success for the RBX2660 2-Dose group (N=25/45; 55.6%) to the Placebo group (N=19/44; 43.2%) in the blinded portion was not statistically significant at the final analysis (p=0.243) in the ITT population (Figure 9). Statistical significance was also not achieved in the sensitivity analyses using the mITT (62.5% vs. 44.2%; p=0.095) and PP (75.0% vs. 58.1%; p=0.170) populations (Table 5).

Regarding the secondary endpoint of regimen-finding, in comparing the 2 RBX2660 treatment arms, no meaningful difference in 1 dose (Treatment Success, 56.8% [25/44]) versus 2 doses (Treatment Success, 55.6% [25/45]) was observed; therefore, a single dose of RBX2660 (rather than 2) was selected for the Pivotal Phase 3 Study.

Figure 9: Phase 2B Study 2014-01 – Primary and Secondary Efficacy Results – 2-Dose RBX2660 Group vs. Placebo Group and 1-Dose RBX2660 Group vs. Placebo Group (ITT)

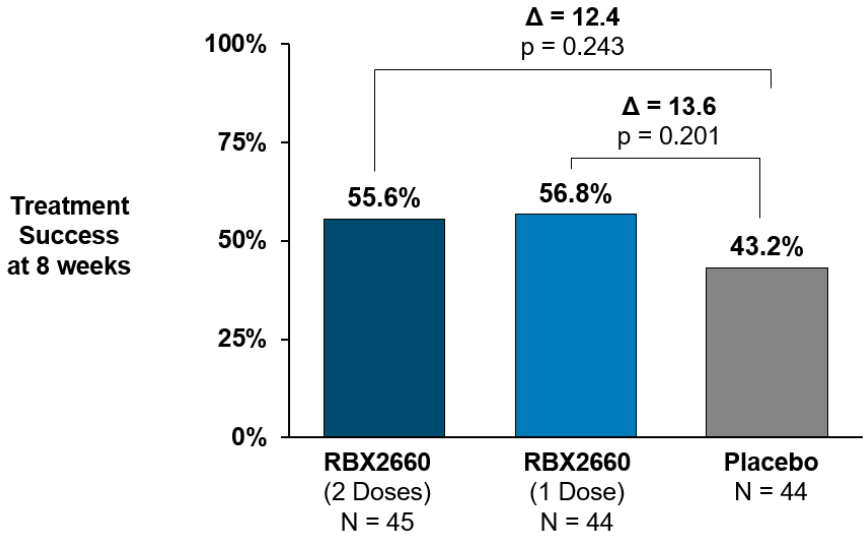


Table 5: Phase 2B Study 2014-01 – Treatment Success 2-Dose RBX2660 Group vs. Placebo Group (ITT, mITT, PP)

	ITT		mITT		PP	
	2-Dose RBX2660 (RBX-RBX) N=45	Placebo (PBO-PBO) N=44	2-Dose RBX2660 (RBX-RBX) N=40	Placebo (PBO-PBO) N=43	2-Dose RBX2660 (RBX-RBX) N=28	Placebo (PBO-PBO) N=31
Success n (%)	25 (55.6)	19 (43.2)	25 (62.5)	19 (44.2)	21 (75.0)	18 (58.1)
Failure n (%)	20 (44.4)	25 (56.8)	15 (37.5)	24 (55.8)	7 (25.0)	13 (41.9)
Failure Indeterminate Untreated	13 (28.9) 3 (6.7) 4 (8.9)	18 (40.9) 7 (15.9) -	12 (30.0) 3 (7.5) -	18 (41.9) 6 (14.0)	7 (25) - -	13 (41.9) - -
Difference [a] 95% CI [b]	12.4 -8.2 to 33.0		18.3 -2.8 to 39.4		16.9 -6.7 to 40.6	
p-value [c]	0.243		0.095		0.170	

[a] Difference in percentage of Treatment Successes between the treatment groups.
[b] Two-sided 95% confidence interval (CI) using normal approximation for difference in percentages between treatments.
[c] P-value from Pearson's chi-square test for the difference between the treatment groups with respect to percentage of Treatment Successes. Indeterminate response was treated as a failure for the purpose of analysis.

6.3.4 Results: Secondary and Other Efficacy Objectives

6.3.4.1 1-Dose RBX2660 Compared to Placebo Group

A secondary efficacy analysis comparing the 1-Dose RBX2660 group (1 dose RBX2660 and 1 placebo dose administered 7 ± 2 days apart) success rate to the Placebo group was conducted (Table 6). In the ITT and mITT populations, the success rate in the 1-Dose RBX2660 group as compared with the Placebo group was not statistically significant (p=0.201 and p=0.051 respectively). However, the PP efficacy analysis comparing the 1-Dose RBX2660 Group success rate of 87.5% to the Placebo group at 58.1% demonstrated a statistically significant difference, with a 29.4%-point difference (p=0.017).

Table 6: Phase 2B Study 2014-01 – Treatment Success in 1-Dose RBX2660 vs. Placebo (ITT, mITT, PP)

	ITT		mITT		PP	
	1-Dose RBX2660 (RBX-PBO) N=44	Placebo (PBO-PBO) N=44	1-Dose RBX2660 (RBX-PBO) N=38	Placebo (PBO-PBO) N=43	1-Dose RBX2660 (RBX-PBO) N=24	Placebo (PBO-PBO) N=31
Success n (%)	25 (56.8)	19 (43.2)	25 (65.8)	19 (44.2)	21 (87.5)	18 (58.1)
Failure n (%)	19 (43.2)	25 (56.8)	13 (34.2)	24 (55.8)	3 (12.5)	13 (41.9)
Failure Indeterminate Untreated	9 (20.5) 8 (18.2) 2 (4.5)	18 (40.9) 7 (15.9) -	9 (23.7) 4 (10.5) -	18 (41.9) 6 (14.0)	3 (12.5) - -	13 (41.9) - -
Difference [a] 95% CI [b]	13.6 -7.1 to 34.3		21.6 0.4 to 42.8		29.4 7.6 to 51.3	
p-value [c]	0.201		0.051		0.017	

[a] Difference in percentage of Treatment Successes between the treatment groups.
[b] Two-sided 95% confidence interval (CI) using normal approximation for difference in percentages between treatments.
[c] P-value from Pearson's chi-square test for the difference between the treatment groups with respect to percentage of Treatment Successes. Indeterminate response was treated as a failure for the purpose of analysis.

6.3.4.2 2-Dose RBX2660 Group Compared to 1-Dose RBX2660 Group

An efficacy analysis was performed comparing the Treatment Success rate of the 2-Dose RBX2660 group (2 doses of RBX2660 administered 7 ± 2 days apart) to the 1-Dose RBX2660 group (Table 7). Results showed no statistically significant difference in Treatment Success in any analysis population between receiving 2 doses versus 1 dose of RBX2660.

Table 7: Phase 2B Study 2014-01 – Treatment Success 2-Dose RBX2660 vs. 1-Dose RBX2660 (ITT, mITT, PP)

	ITT		mITT		PP	
	2-Dose RBX2660 (RBX-RBX) N=45	**1-Dose RBX2660 (RBX-PBO) N=44**	**2-Dose RBX2660 (RBX-RBX) N=40**	**1-Dose RBX2660 (RBX-PBO) N=38**	**2-Dose RBX2660 (RBX-RBX) N=28**	**1-Dose RBX2660 (RBX-PBO) N=24**
Success n (%)	25 (55.6)	25 (56.8)	25 (62.5)	25 (65.8)	21 (75.0)	21 (87.5)
Failure n (%)	20 (44.4)	19 (43.2)	15 (37.5)	13 (34.2)	7 (25.0)	3 (12.5)
Failure	13 (28.9)	9 (20.5)	12 (30.0)	9 (23.7)	7 (25.0)	3 (12.5)
Indeterminate	3 (6.7)	8 (18.2)	3 (7.5)	4 (10.5)	-	-
Untreated	4 (8.9)	2 (4.5)	-	-	-	-
Difference [a]	1.3		3.3		12.5	
95% CI [b]	-19.4 to 21.9		-18.0 to 24.6		-8.3 to 33.3	
p-value [c]	0.904		0.762		0.254	

[a] Difference in percentage of Treatment Successes between the treatment groups.
[b] Two-sided 95% CI using normal approximation for difference in percentages between treatments.
[c] P-value from Pearson's chi-square test for the difference between the treatment groups with respect to percentage of Treatment Successes. Indeterminate response was treated as a failure for the purpose of analysis.

6.3.4.3 Treatment Results After Second Course of RBX2660

Study 2014-01 allowed for a second, open-label course of RBX2660 treatment for patients with laboratory-confirmed Treatment Failure within the first 8 weeks after treatment. Among the 19 patients with Treatment Failure in the 1-dose RBX2660 group, 14 patients were eligible and opted for a second course of treatment. More than half of these patients (57.1%; 8/14) reported Treatment Success after the additional 8 weeks following the second course of treatment.

6.4 Pivotal Phase 3 Study 2017-01

6.4.1 Investigational Plan

6.4.1.1 Overall Design

Pivotal Study 2017-01 was a prospective, multicenter, randomized, double-blinded, placebo-controlled Phase 3 study to confirm the efficacy and evaluate the safety of RBX2660 for the prevention of rCDI. A schematic of the study design is shown in Figure 2.

Up to 270 patients were to be randomized and treated in the study. Randomization was at a 2:1 ratio (RBX2660:Placebo); approximately 180 patients were to be randomized to RBX2660 treatment and 90 patients randomized to placebo.

Patients who were deemed Treatment Failures following the blinded treatment, per the prespecified Treatment Failure definition, could choose to receive an open-label RBX2660 dose.

A primary efficacy analysis and safety analyses were conducted when all randomized patients had completed their 8-week follow-up assessment after receiving the blinded treatment. Two

interim analyses were conducted to evaluate the primary efficacy endpoint for possible early study stopping for success or futility. The first interim analysis could occur after a minimum of 160 patients were treated and evaluated for efficacy. A second interim analysis could occur when 220 patients were treated and evaluated for efficacy if success was not achieved at the first interim analysis.

6.4.1.1.1 Definitions of Treatment Success, Sustained Clinical Response, and Treatment Failure

Definitions

Treatment Success was defined as:

- The absence of CDI diarrhea (passage of 3 or more unformed stools in 24 or fewer consecutive hours for at least 2 consecutive days and a positive stool test for *C. difficile* toxin documented at the time of diarrhea) for 8 weeks after completing a study treatment.

Sustained Clinical Response was defined as:

- Treatment Success of the presenting CDI recurrence and no new CDI episodes for greater than 8 weeks through 6 months after completing a study treatment.

Treatment Failure was defined as:

- The presence of CDI diarrhea within 8 weeks of administration of a study dose, which included a positive stool test for *C. difficile* toxin at the time of the diarrhea.

Determination of Success/Failure (Investigator and Endpoint Adjudication Committee)

The site investigator made the initial determination of success or failure based on the predefined study definitions. The site investigator's assessment as well as a data package for each patient was then provided to the chartered Endpoint Adjudication Committee (EAC) for independent, blinded adjudication of treatment outcome that was utilized for study analysis and reporting purposes.

Management of Treatment Failures

Following blinded treatment, patients with Treatment Failure could be scheduled for administration of an open-label RBX2660 dose within 21 calendar days of failure determination.

The use of antibiotics prior to an open-label RBX2660 dose was at the discretion of the investigator. If antibiotics were given to control symptoms, a 24-72 hour washout period prior to administration of an open-label RBX2660 dose was required

If a patient received an open-label RBX2660 dose, the follow-up requirements restarted from the day of administration of the open-label RBX2660 dose according to the same schedule as required for the blinded portion of the study.

Treatment Failures who did not receive an open-label RBX2660 dose were to continue to follow their original schedule of assessments for the duration of the study based on the blinded study dose.

6.4.1.2 Randomization and Treatments

For the blinded treatment, the randomization schedule was created using randomized blocks within 4 strata, based on antibiotics used at screening (Vancomycin alone, Vancomycin in

combination, Fidaxomicin, or Other), and randomized patients were assigned to one of 2 treatments:

- 1 dose of RBX2660
- 1 dose of placebo

Active treatment was RBX2660 (fecal microbiota suspension) in a dose of 150 mL supplied in an enema bag.

The placebo enema consisted of 150 mL of normal saline.

Duration of Treatment

Patients were expected to participate in the study for approximately 7 months: 1 month enrollment and 6 months follow-up.

6.4.1.3 Objectives and Endpoints

6.4.1.3.1 Primary and Secondary Objectives

Primary objective: To confirm the efficacy of RBX2660 as compared to a placebo in preventing recurrent episodes of CDI through 8 weeks.

Secondary objective: To evaluate the Sustained Clinical Response rate of RBX2660 as compared to placebo after blinded treatment.

6.4.1.3.2 Primary and Secondary Endpoints

The primary efficacy endpoint was Treatment Success, i.e., the absence of CDI diarrhea for 8 weeks after completing the blinded study treatment in the mITT population. The primary analysis of the study was a Bayesian hierarchical model, which formally incorporated data from the Phase 2B study (Study 2014-01). This analysis tested the hypothesis that the success rate of RBX2660 was superior to placebo.

The secondary endpoint was Sustained Clinical Response through 6 months after blinded treatment.

6.4.1.3.3 Other Endpoints

1. Baseline characteristics
2. Patient fecal microbial composition at Screening, and 4 weeks, 8 weeks, 3 months, and 6 months after blinded study treatment
3. Charlson Comorbidity Index at Screening, 8-week, 3-, and 6-month phone assessments
4. Health-related quality of life (per Cdiff32 questionnaire) at Screening, Week 1, 4, 8, month 3 and 6
5. ATLAS score (calculated from Age, Temperature, Leucocytes, Albumin, and Systematic antibiotics) for CDI severity of qualifying CDI event
6. Recurrence of CDI within 8 weeks of open-label RBX2660 treatment in placebo patients who were documented blinded study Treatment Failures
7. Recurrence of CDI within 8 weeks of blinded or open-label RBX2660 treatment
8. Occurrence of CDI through 6 months in all patients receiving a single dose of RBX2660

9. Concentration of vancomycin-resistant *Enterococcus* in stool samples for patients who were carriers at baseline
10. Presence of *C. difficile* in stool samples at Screening, and 4 weeks, 8 weeks, 3 months and 6 months after study treatment

6.4.1.4 Selection of Study Population

The most significant difference in eligibility criteria between the Phase 2B Study 2014-01 and the Pivotal Phase 3 Study 2017-01 was that, while the Phase 2B study enrolled only patients with ≥ 2 recurrences of CDI and ≥ 2 rounds of standard of care oral antibiotic therapy, the Phase 3 study required only 1 recurrence of CDI and ≥ 1 round of standard of care oral antibiotic therapy for enrollment. Additional details on eligibility criteria for the Pivotal Phase 3 study are provided below.

Inclusion criteria for Pivotal Phase 3 Study 2017-01 were:

1. Adult patients ≥ 18 years old.
2. Medical record documentation of rCDI per the study definition, including either: a) at least 1 recurrence after a primary episode and had completed at least 1 round of standard of care oral antibiotic therapy or b) had at least 2 episodes of severe CDI resulting in hospitalization within the last year.
3. A positive stool test for the presence of toxigenic *C. difficile* within 30 days prior to enrollment.
4. Was currently taking or had just been prescribed antibiotics to control CDI-related diarrhea at the time of enrollment. Note: Patient's CDI diarrhea had to be controlled (<3 unformed/loose, ie, Bristol Stool Scale type 6-7, stools/day for 2 consecutive days) while taking antibiotics during screening.
5. Was willing and able to have an enema(s).
6. Was willing and able to complete the stool and serum testing required for the study.
7. Agreed not to take non-dietary probiotics through 8 weeks after receiving the last study enema (including over-the-counter and prescription).
8. Agreed not to take any oral vancomycin, metronidazole, fidaxomicin, rifaximin, nitazoxanide, and intravenous immunoglobulin through the 8-week follow-up assessment unless newly prescribed by a treating investigator during the course of the study as a result of rCDI diagnosis.
9. Agreed to practice a form of effective contraception during study participation; did not apply to persons with documented non-childbearing potential.
10. Had a negative urine pregnancy test at the time of enrollment and on the day of each enema prior to administration (persons of childbearing potential only).
11. Was willing and able to provide informed consent and local privacy authorization as applicable.
12. Was willing and able to complete the required Patient Diary.

13. Was willing and able to meet all study requirements, including attending all assessment visits and telephone calls.

Exclusion criteria included the following:

1. A known history of refractory CDI.
2. Had continued CDI diarrhea despite being on a course of antibiotics prescribed for CDI treatment.
3. Required antibiotic therapy for a condition other than CDI.
4. Previous FMT, RBX2660 treatment, receipt of CDI vaccine, or treatment with CDI monoclonal antibodies prior to study enrollment.
5. History of IBD, eg, ulcerative colitis, Crohn's disease, or microscopic colitis.
6. Diagnosis of IBS as determined by Rome III criteria.
7. History of chronic diarrhea.
8. History of celiac disease.
9. Disease symptoms (diarrhea) caused by a confirmed intestinal pathogen other than *C. difficile*.
10. Currently had a colostomy.
11. Intraabdominal surgery within the last 60 days.
12. Evidence of active, severe colitis.
13. History of short gut syndrome or motility disorders.
14. Required the regular use of medications to manage bowel hypermotility.
15. Had planned therapy within 3 months that might cause diarrhea (eg, chemotherapy).
16. Planned surgery requiring perioperative antibiotics within 6 months of study enrollment.
17. Life expectancy of < 6 months.
18. Compromised immune system (eg, Human immunodeficiency virus (HIV) infection with a CD4 count <200/mm3; inherited/primary immune disorders; immunodeficient or immunosuppressed due to a medical condition or medication). Note: Eligible HIV patients who had a CD4 count >200/mm3 who were on stable, highly active anti-retroviral therapy were considered for enrollment.
19. Taking systemic steroids > 20 mg prednisone a day or prednisone-equivalent or was expected to be on steroids (> 20 mg prednisone a day or equivalent) after enrollment through 8 weeks after completing the assigned study treatment. Note: Eligible patients taking a steroid dose equivalent to prednisone 20 mg/day for >2 weeks, antimetabolites (eg, azathioprine, 6-mercaptopurine, or low-dose methotrexate for autoimmune disease), calcineurin inhibitors (eg, tacrolimus and cyclosporine), or mycophenolate mofetil may have been enrolled only after consultation with the Medical Monitor, and only if the doses had been stable (except for drug therapeutic monitoring adjustments for calcineurin inhibitors) for 90 days and had not been associated with diarrhea prior to the current episode of CDI.

20. An absolute neutrophil count of <1000 cells/μL during screening.
21. Known or suspected current (< 90 days) illicit drug use. Note: marijuana use was allowed.
22. Participant was unable to discontinue opioids (unless on a stable dose with no increase in dose planned for the duration of the study). *Note: Opioids were permitted as needed as long as participants were on a stable dose at the time of randomization and expect to maintain the same dose until the 8 week follow-up visit OR if the participant had been on short-term (i.e., ≤ 14 days) opioid treatment and there was anticipation of a dose decrease or cessation of use during the course of the study. Participants who only received a few doses at the time of presentation of CDI could be considered for participation. Investigator was to consult on any clarification of the opioid doses/treatment with the Medical Monitor.*
23. Pregnant, breastfeeding, or intended to become pregnant during study participation.
24. Participating in a clinical trial of another investigational product (drug, device or other) and had not completed the required follow-up period.
25. Patient, in the opinion of the investigator, for whatever reason, should have been excluded from the study.

6.4.1.5 Statistical Analysis

6.4.1.5.1 Data Sets Analyzed

There were 4 analysis populations:

- **Safety Population (SP):** The SP was defined as the population of randomized patients who received any study treatment. Patients were analyzed according to the treatment they actually received.
- **Intent-to-Treat Population (ITT):** All randomized patients. Patients were analyzed according to the randomized treatment rather than the actual treatment received regardless of treatment misallocations. Randomized patients who exited prior to receiving blinded treatment were not included in the analysis.
- **Modified Intent-to-Treat Population (mITT):** All randomized patients who successfully received blinded treatment but excluding patients who withdrew prior to treatment; patients in whom treatment was attempted but not completed; and patients who discontinued from the study prior to evaluation of Treatment Failure or Treatment Success for the primary endpoint if the reason for exit was not related to CDI symptoms.
- **Per Protocol Population (PP):** All patients who successfully received blinded treatment and were analyzed according to the treatment they received, excluding patients who had documented deviations to inclusion or exclusion criteria; and patients who exited the study prior to the 8-week efficacy evaluation if the reason for exit was not related to CDI symptoms.

6.4.1.5.2 Analyses of the Primary Efficacy Endpoint

Study 2017-01 used a Bayesian adaptive design to evaluate the efficacy of RBX2660 on the primary efficacy endpoint, the recurrence of CDI within 8 weeks of blinded treatment. This design incorporated both dynamic borrowing of information from the prior randomized placebo-controlled Phase 2 study, 2014-01, as well as interim analyses to determine whether the trial could stop

early for futility or overwhelming evidence of efficacy. As described earlier (Section 4.2), this approach was taken at the recommendation of FDA due to recruitment difficulties during the clinical program with the goal of allowing the single adequate and well-controlled Phase 3 trial, 2017-01, to serve as substantial evidence of effectiveness to support the approval of RBX2660.

The planned primary analysis for Study 2017-01 was based on the mITT population, with planned sensitivity analyses for the ITT and PP populations. At the time when the Bayesian hierarchical model was incorporated into the prospective analysis of the primary endpoint in Study 2017-01, the results for Study 2014-01 had already been preliminarily reported. As the ITT population was the primary analysis population in Study 2014-01, it was decided that the dynamic borrowing should be based on the 2014-01 ITT analysis population. Following advice from FDA, only data from the 1-Dose RBX2660 group and the Placebo group was borrowed. In all planned analyses, borrowing was based on the 2014-01 ITT analysis population. A complete technical description of the Bayesian adaptive design and statistical modeling can be found in Appendix 11.1.

During BLA review, FDA pointed out that aligning the analysis populations and definitions would lead to a stronger claim of exchangeability between Studies 2014-01 and 2017-01. Therefore, an updated primary efficacy endpoint analysis was performed using the Bayesian hierarchical model, further aligning these studies for borrowing by applying the Study 2017-01 definitions of the analysis populations to the Study 2014-01 efficacy data, matching populations when borrowing, and restricting the follow-up period in Study 2014-01 to 8 weeks from first dose.

Brief Background on Frequentist and Bayesian Inference

Traditional frequentist statistical tests (eg, t-tests, chi-square tests) evaluate hypotheses using only observed data from a single study. Statistical significance is determined using a p-value, which describes the likelihood of the observed results or more extreme, assuming that the null hypothesis is true. For example, consider a study with a treatment group and a control group that are compared using a 1-sided test for superiority. A p-value of 0.01 would mean that there is a 1% probability of observing a treatment difference at least as large as observed in the study by chance alone if, in truth, the treatment was actually identical to the control.

In contrast to frequentist methods, Bayesian inference involves integrating both observed data as well as prior information. Prior information could include the results of a prior study, multiple prior studies, or expert opinion; the weighting of the prior information relative to the new evidence can be data-driven or explicitly selected. Statistical significance is determined using the posterior probability, which directly describes the probability of interest (in clinical trials, typically the probability that the treatment is superior to the control). Taking the same example of a study with a treatment group and a control group and a 1-sided test for superiority, a posterior probability of 0.99 means that the probability that the treatment group is superior to the control group is 99%.

In both the frequentist and Bayesian examples, the statistical conclusion about the treatment being superior to control is the same; however, in the Bayesian test, we do not need to assume the null hypothesis is true to conduct statistical inference, and the frequentist test cannot incorporate prior information.

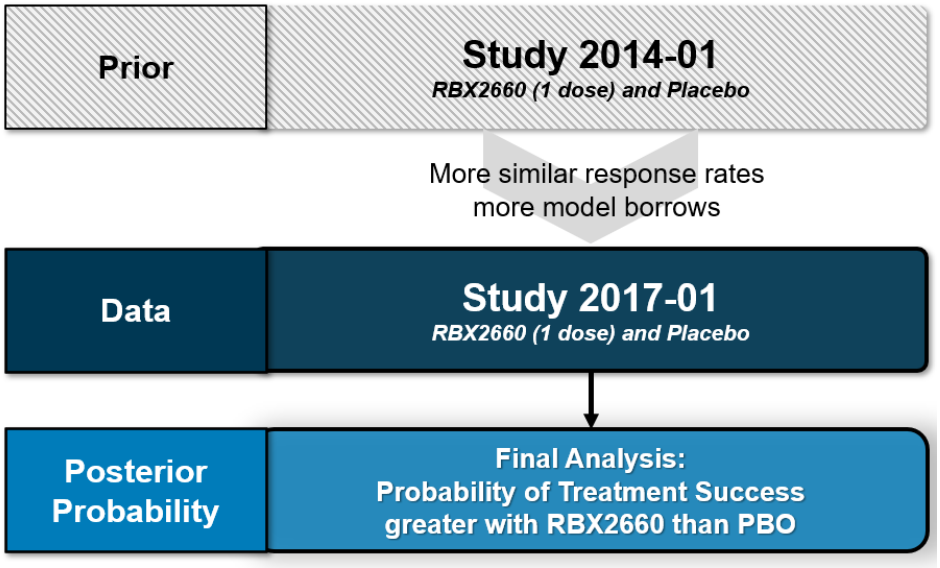
Dynamic Borrowing and Hierarchical Model

The primary analysis of the primary efficacy endpoint was conducted using a Bayesian hierarchical model. This model allowed for the dynamic borrowing of information about the

treatment and control effects from Study 2014-01 into the evaluation of Study 2017-01. (See Appendix 11.1 for details about the model parameterization.)

The patients in Study 2017-01 were enrolled from a similar population to Study 2014-01. However, to ensure that an appropriate amount of information was borrowed from the Phase 2 study, a dynamic borrowing approach was utilized to account for potential heterogeneity between the study populations. If the observed data from the studies were similar, the prior data from Study 2014-01 would contribute more information to the final inference (Figure 10). On the other hand, if the newly observed data in Study 2017-01 were quite different from Study 2014-01, the prior data would contribute less information to the inference. Thus, the degree of borrowing of information from 2014-01 into the final analysis of 2017-01 was driven by the data, rather than being specified in advance. Only the data from the 1-dose RBX2660 group and Placebo group from Study 2014-01 were utilized for the Bayesian modeling.

Figure 10: Pivotal Phase 3 Study 2017-01 - Bayesian Hierarchical Model



Success Criteria and Futility Stopping

The maximum sample size for Study 2017-01 was set at 270 patients, however the design allowed for 2 interim analyses at enrollment targets of 160 and 220 patients, respectively, to stop the study early – either for futility or for overwhelming evidence of efficacy. Both interim analyses and the final analysis were performed by a Statistical Analysis Committee independent of the Sponsor, DSMB, and EAC.

Study 2017-01 was to stop for futility at either interim analysis if the predictive probability of trial success if enrollment continued up to the maximum sample size was less than 0.01.

The success criteria at each analysis were predicated on the treatment effect of RBX2660 relative to placebo. The statistical significance of the treatment effect was evaluated by calculating the posterior probability of superiority for the RBX2660 group versus the Placebo group. Type I error due to the interims was controlled by utilizing a Pocock spending function for the 2 interim analyses and a final analysis with a cumulative alpha spend of 0.00125.

After adjusting for Type I error spending, the success criterion for the posterior probability at the interim analyses and the final analysis was set at 99.93% probability that RBX2660 was superior

to control. The FDA agreed that demonstrating statistical significance at this level would be considered "a statistically very persuasive finding" that would constitute substantial evidence of effectiveness in lieu of 2 positive adequate and well-controlled trials.

At the final analysis, a second threshold of 97.50% allowed for success to be declared at the 0.025 alpha level. This lower threshold controlled the overall Type I error rate for the trial at 2.5% accounting for the alpha spending at the interim and final analyses. Thus, achieving a posterior probability of at least 97.50% but less than 99.93% at the final analysis would be considered equivalent to a single positive adequate and well-controlled trial (Figure 11).

Figure 11: Pivotal Phase 3 Study 2017-01 - Statistical Significance Thresholds

	Bayesian Posterior Probability Superiority
Statistically significant	**97.50%**
Statistically very persuasive	**99.93%**

6.4.1.5.3 Analyses of the Secondary Endpoint

The rates of Sustained Clinical Response (ie, the rate of Treatment Success at 8 weeks of the presenting CDI recurrence and no new CDI episodes for up to 6 months after completing a blinded study treatment) were compared between the RBX2660 group and the control group using a chi-square test. Patients who exited prior to their 6-month follow-up were conservatively counted as a Treatment Failure.

A hierarchical, closed-testing procedure was utilized for the secondary endpoint such that the test would only be performed if the primary efficacy endpoint was met. The secondary endpoint was to be tested at the 2-sided 0.05 significance level.

6.4.1.5.4 Handling of Missing Data for the Primary Endpoint

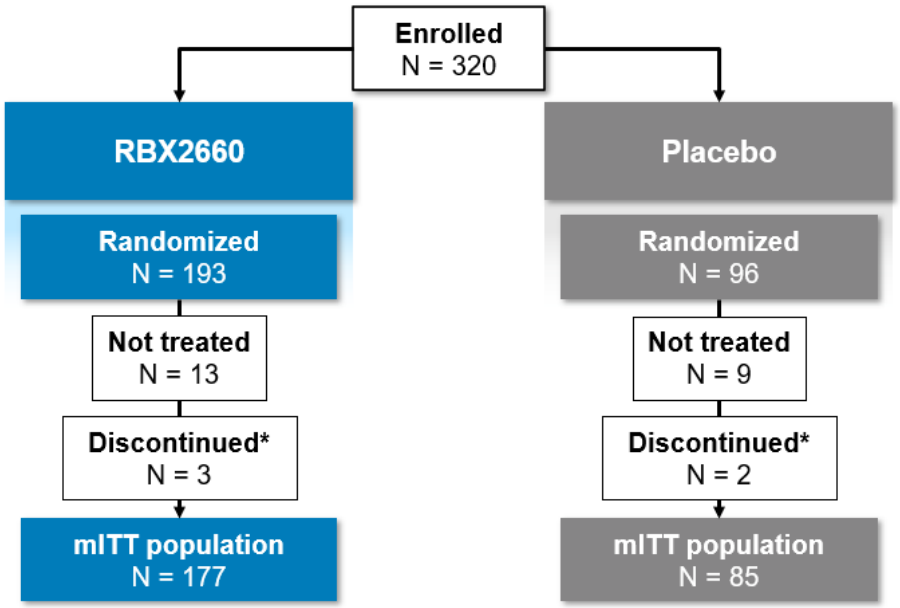
Patients who exited prior to the 8-week visit without an assessment of Treatment Success or Failure were considered Treatment Failures.

6.4.2 Study Patients

6.4.2.1 Disposition

Of the 320 patients that enrolled, 289 were randomized: 193 to RBX2660 and 96 to placebo. A total of 267 were treated, and 5 discontinued prior to the 8-week efficacy analysis due to non-CDI-related symptoms. Therefore, 262 patients are included in the mITT population: 177 treated with RBX2660 and 85 with placebo (Figure 12).

Figure 12: Pivotal Phase 3 Study 2017-01 – Patient Disposition in mITT Population



6.4.2.2 Baseline Demographics and Characteristics

There were no notable differences in the baseline demographics between the treatment arms (Table 8). Baseline demographic characteristics were generally representative of an adult patient population with rCDI. The mean ages of the patients were approximately 61 years in the RBX2660 group and 58 years in the Placebo group, the majority of patients were female and mostly White.

Table 8: Pivotal Phase 3 Study 2017-01 - Baseline Demographics and Characteristics (mITT Analysis Set)

	RBX2660 N = 177	**Placebo N = 85**
Age (years), mean (SD) Min, max	61.3 (16.8) 19-93	57.5 (15.9) 26-86
Female, n (%)	122 (68.9%)	59 (69.4%)
White, n (%)	165 (93.2%)	76 (89.4%)
Duration of CDI (days), mean (SD)	26.3 (14.8)	25.3 (11.4)
Previous episodes of CDI*, mean	3	3
Hospitalization		
Due to CDI episode, n (%)	23 (13.0%)	10 (11.8%)
Duration (days), median (IQR)	5.0 (4.0)	5.0 (4.0)
Vancomycin during screening, n (%)	154 (87.0%)	76 (89.4%)

*Prior to study entry

6.4.3 Results: Primary Efficacy Endpoint

6.4.3.1 Primary Efficacy Endpoint – Initial Bayesian Analysis

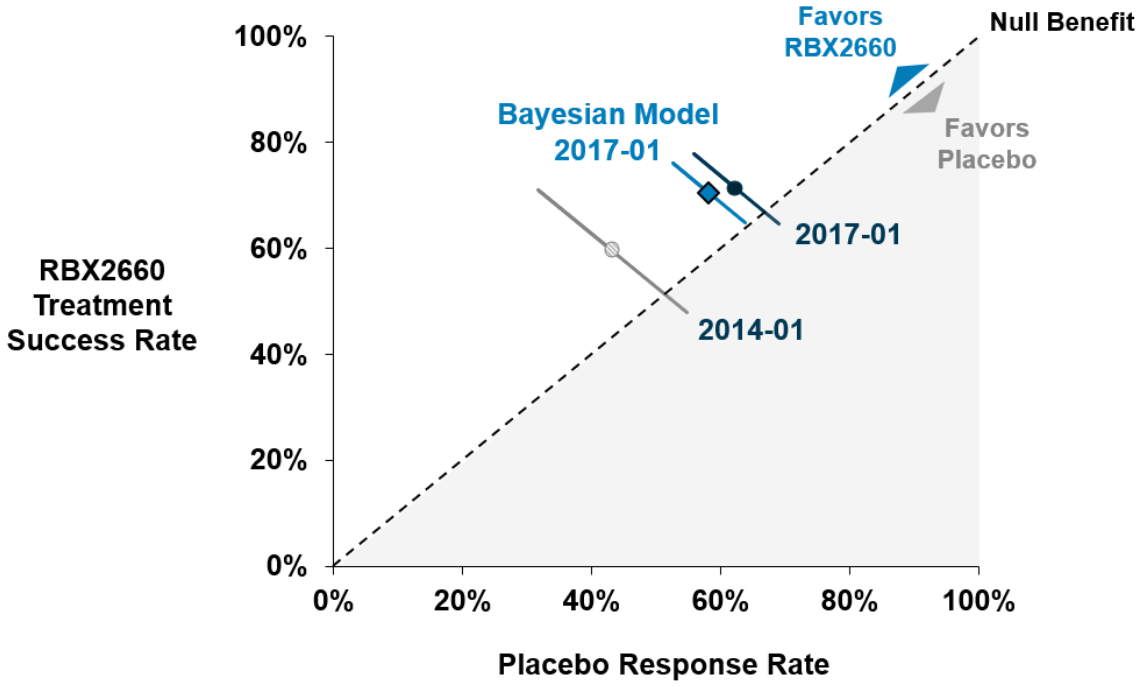
At the final analysis, RBX2660 was superior to placebo in the prevention of CDI recurrence through 8 weeks of blinded treatment. Per the originally planned Bayesian analysis, the model-

estimated Treatment Success rate was 70.4% in the RBX2660 group and 58.1% in the Placebo group. The difference in Treatment Success rates was 12.3 percentage points (95% credible interval [CrI]: 1.4 to 23.3). The probability that RBX2660 was superior to placebo was 98.6%. Thus, the study surpassed the success criterion of 97.5%, but not the higher threshold of 99.9%.

The sensitivity analyses of the primary endpoint using the Bayesian hierarchical model in the ITT and PP populations showed results consistent with the primary analysis. In the ITT population, model-estimated Treatment Success rates were 69.1% and 56.7% in the RBX2660 and Placebo groups, respectively, with a 12.5 percentage point treatment difference (95% CrI: 1.6 to 23.3) and a 98.7% posterior probability that RBX2660 was superior to placebo. In the PP population, model-estimated Treatment Success rates were 70.9% and 57.2% in the RBX2660 and Placebo groups, respectively, with a 13.7 percentage point treatment difference (95% CrI: 2.4 to 25.1) and a 99.1% posterior probability that RBX2660 was superior to placebo.

Figure 13 illustrates the Treatment Success rates for RBX2660 and placebo in the individual trials and the outcome of the Bayesian hierarchical model. The figure shows the RBX2660 response rate on the y-axis and the placebo response rate on the x-axis. The diagonal line equals null benefit, with values above the line corresponding to a superior response rate for RBX2660 compared to placebo. The treatment differences and 95% confidence intervals based on independent analysis of the individual trials are shown. The Bayesian model treatment difference and credible interval are shown in light blue and show that the Bayesian credible interval do not cross the diagonal null benefit line.

Figure 13: Pivotal Phase 3 Study 2017-01 – Bayesian Hierarchical Model Results (mITT)



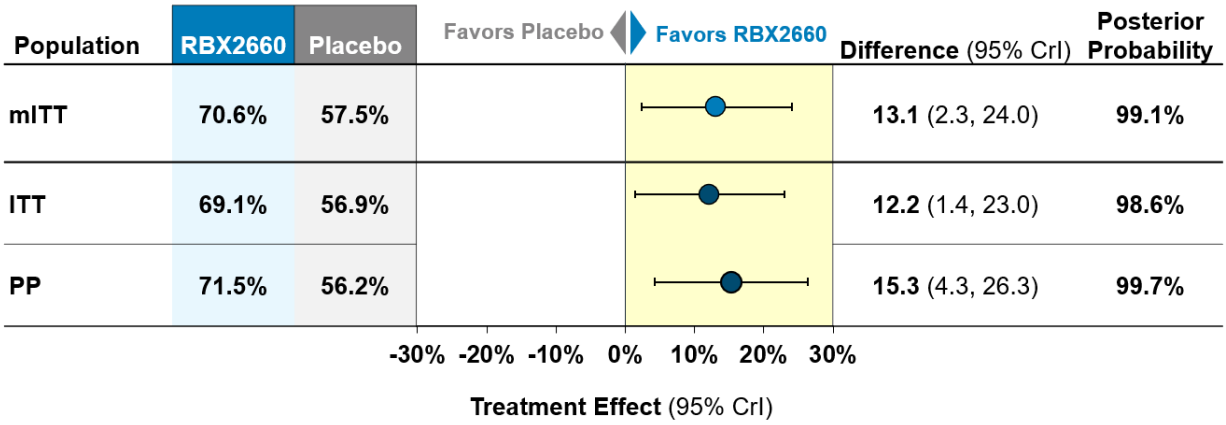
6.4.3.2 *Primary Efficacy Endpoint – Bayesian Analysis Requested by FDA During BLA Review*

During the BLA review process, FDA requested additional Bayesian analyses to further support exchangeability between Studies 2014-01 and 2017-01. As described in Section 4.2, Study 2017-01 was not originally designed to utilize a Bayesian hierarchical model to dynamically borrow information from the prior randomized placebo-controlled Phase 2B Study 2014-01. Therefore, some differences exist between the two protocols and can be summarized as follows:

- The definitions of the analysis populations were not identical.
- In both trials, Treatment Success was defined as absence of CDI recurrence within 8 weeks of completing treatment. However, since Study 2014-01 investigated a dosing regimen with 2 doses administered 1 week apart, the follow-up period from the first dose was effectively 9 weeks.

When the Bayesian hierarchical model was incorporated into the prospective analysis of the primary endpoint in Study 2017-01, the results for Study 2014-01 had already been preliminarily reported. It was therefore decided to borrow the pre-planned primary analysis results from the Study 2014-01 ITT population. During BLA review, FDA pointed out that aligning the analysis population definitions would lead to a stronger claim of exchangeability between Studies 2014-01 and 2017-01. Figure 14 summarizes the results of the Bayesian hierarchical model, which align these studies for borrowing by applying the Study 2017-01 definitions of the analysis populations to the Study 2014-01 efficacy data, matching populations when borrowing, and restricting the follow-up period in Study 2014-01 to 8 weeks from first dose.

Figure 14: Pivotal Phase 3 Study 2017-01 – Bayesian Analysis Requested by FDA During BLA Review



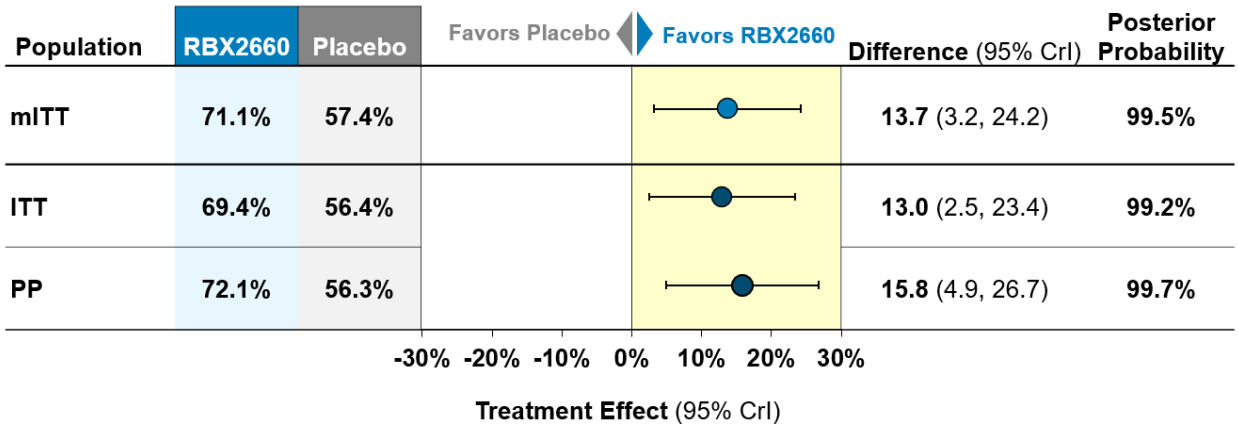
6.4.3.3 *Primary Efficacy Endpoint – Bayesian Sensitivity Analysis Requested by FDA During BLA Review*

In addition to the differences noted in the previous section, the Phase 2B and Phase 3 trials also differed with respect to inclusion criteria #2:

- Study 2014-01 required at least 2 recurrences after a primary episode, while Study 2017-01 required at least 1 recurrence after a primary episode.

During the BLA review process, FDA requested a sensitivity analysis aligning the definitions as outlined in Section 6.4.3.2 above and also including number of prior CDI episodes as a patient-level covariate in the Bayesian hierarchical model. The result of this analysis is summarized in Figure 15.

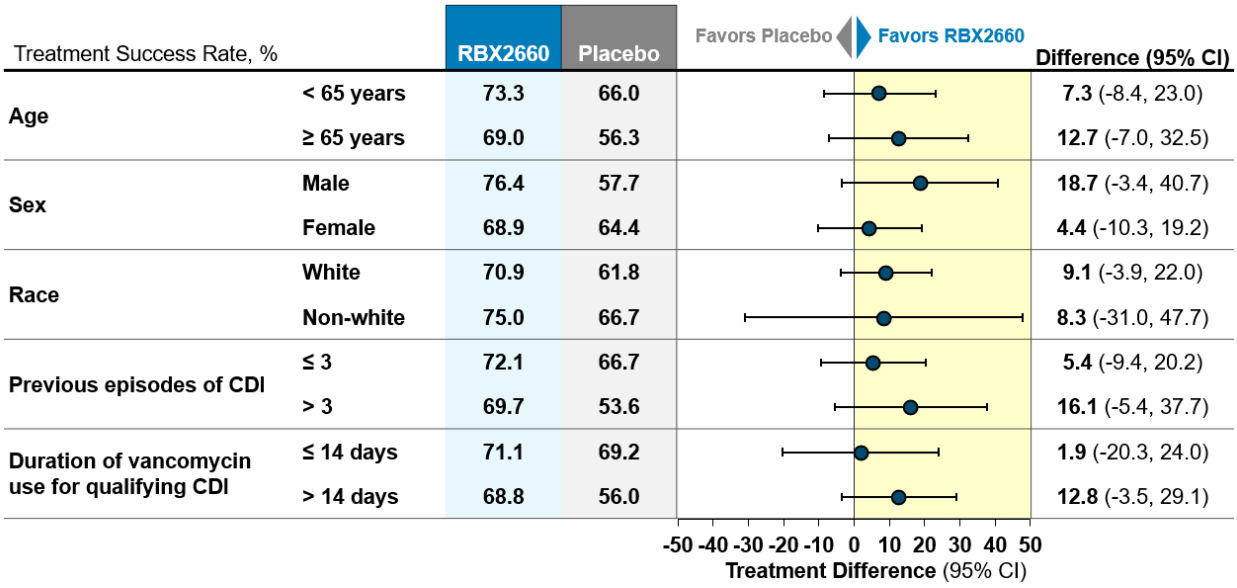
Figure 15: Pivotal Phase 3 Study 2017-01 – Bayesian Sensitivity Analysis Requested by FDA During BLA Review



6.4.3.4 *Primary Efficacy Endpoint – Planned Subgroup Analyses*

Treatment effect was generally consistent with the primary analysis across subgroups (Figure 16). It should be noted that subgroup analyses were performed without the Bayesian methodology that was used for the overall primary efficacy analysis.

Figure 16: Pivotal Phase 3 Study 2017-01 – Primary Efficacy Results in Subgroup Analyses



6.4.4 Results: Secondary Endpoint – Sustained Clinical Response

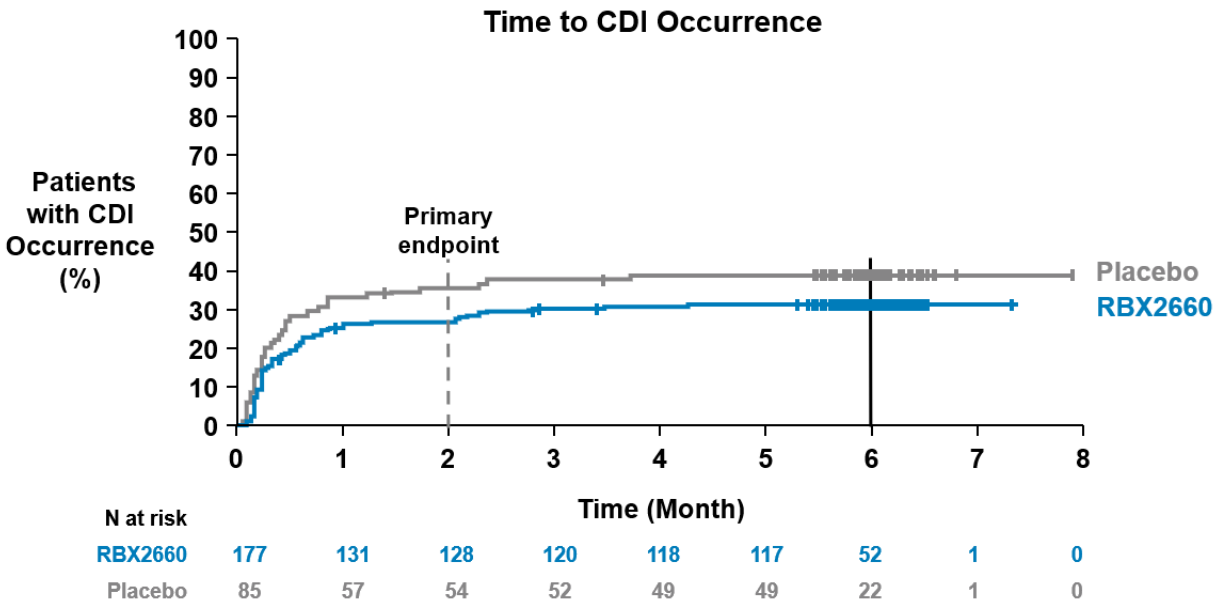
The observed Treatment Success and Treatment Failure rates through 8 weeks and 6 months of follow-up following blinded treatment are shown in Table 9. The observed treatment difference at 8 weeks was maintained at 6 months across all analysis populations. The proportion of patients with Treatment Success at 8 weeks that remained free of CDI occurrences was around 90% for both treatment groups across analysis populations. This is further illustrated in the Kaplan-Meier plot in Figure 17 showing the percentage of patients reporting a CDI event over 6 months of follow-up from start of blinded treatment. Compared with placebo, the lower Treatment Failure rate observed for RBX2660 at 8 weeks was also maintained through 6 months of follow-up.

Table 9: Pivotal Phase 3 Study 2017-01 - Observed Treatment Response Rates at 8 weeks and 6 months (mITT)

	ITT		mITT		PP	
	RBX2660 N=180	**Placebo N=87**	**RBX2660 N=177**	**Placebo N=85**	**RBX2660 N=167**	**Placebo N=78**
Through 8 weeks						
Success, n (%)	126 (70.0)	53 (60.9)	126 (71.2)	53 (62.4)	120 (71.9)	48 (61.5)
Failure, n (%)	54 (30.0)	34 (39.1)	51 (28.8)	32 (37.7)	47 (28.1)	30 (38.5)
Difference [a] 95% CI [b]	9.1 -3.2 to 21.3		8.8 -3.4 to 21.1		10.3 -2.5 to 23.1	
p-value [c]	0.139		0.150		0.105	
Through 6 months						
Success, n (%)	116 (64.4)	48 (55.2)	116 (65.5)	48 (56.5)	110 (65.9)	43 (55.1)
Failure, n (%)	64 (35.6)	39 (44.8)	61 (34.5)	37 (43.5)	57 (34.1)	35 (44.9)
Difference [a] 95% CI [b]	9.3 -3.3 to 21.9		9.1 -3.6 to 21.7		10.7 -2.4 to 23.9	
p-value [c]	0.145		0.156		0.106	
From 8 weeks through 6 months[d]						
Yes, n (%)	116 (92.1)	48 (90.6)	116 (92.1)	48 (90.6)	110 (91.7)	43 (89.6)
No, n (%)	10 (7.9)	5 (9.4)	10 (7.9)	5 (9.4)	10 (8.3)	5 (10.4)

[a] Difference in percentage of Treatment Successes between the treatment groups
[b] Two-sided 95% confidence interval (CI) using the normal approximation for difference in percentages between treatments
[c] p-value from Pearson's chi-square test for the difference between treatment groups with respect to the percentage of Treatment Successes.
[d] The denominator is the number of patients with Treatment Success at 8 weeks

Figure 17: Pivotal Phase 3 Study 2017-01 – Kaplan-Meier Curve of Time to CDI Occurrence by Treatment Group (mITT)



6.4.5 Results: Other Efficacy Endpoints

Results for other efficacy endpoints (listed in Section 6.4.1.3.3) were generally supportive of the primary efficacy results; additional details are provided in Appendix 11.2 in Table 29.

6.4.6 Treatment Results After Second Course of RBX2660

Patients who had Treatment Failure within the first 8 weeks (whether they received RBX2660 or placebo) were offered a second treatment with open-label RBX2660. Among the 51 eligible patients who had been treated with RBX2660 during the blinded period of the study but had Treatment Failure, 41 eligible patients chose to receive a second treatment with open-label RBX2660, and more than half (22/41; 53.7%) had absence of CDI recurrence within 8 weeks of completing treatment (mITT population).

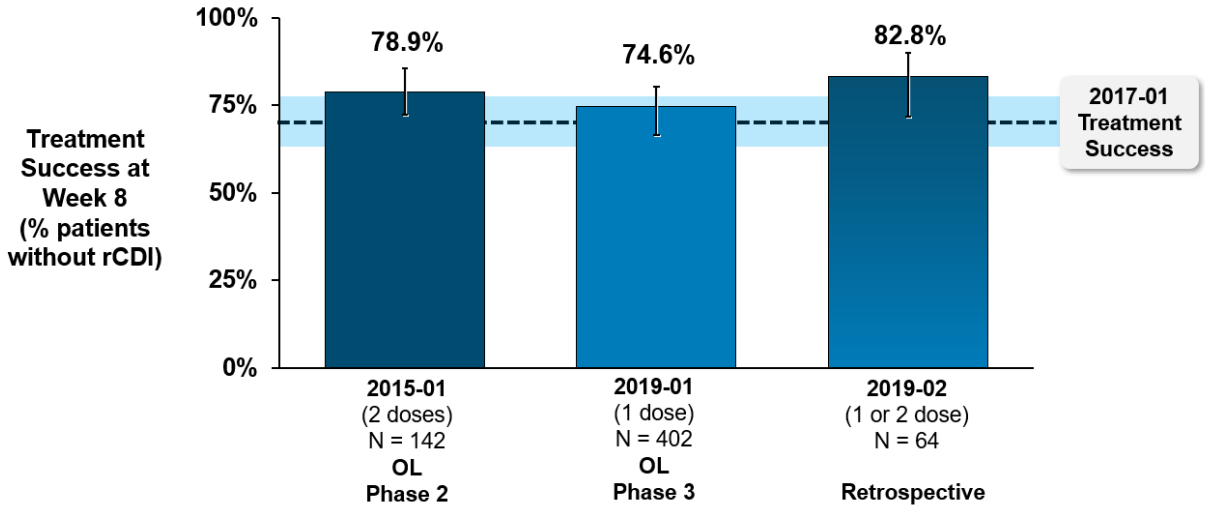
6.5 Supportive Efficacy Results from Open-Label Studies

Study designs for the prospective open-label studies were similar to 2014-01 and 2017-01. Studies 2019-01 and 2019-02 included patients with comorbidities commonly seen among patients with CDI, such as IBS or IBD. Since 2019-02 was a retrospective study, dosing was at the discretion of the treating physician. Follow-up durations for all 4 studies ranged from 6 to 24 months (Table 2).

The incidence of rCDI was comparable across the open-label studies and also comparable with the Pivotal Phase 3 Study 2017-01 (Figure 18). Enrollment criteria became less restrictive in 2019-01, allowing patients with common comorbidities, such as IBD and IBS, to participate; Treatment Success remained high at 74.6%. This pattern continued in the retrospective Study 2019-02, where a total of 82.8% of patients had Treatment Success with either 1 or 2 doses of RBX2660 for a single rCDI event. These results reflect use in clinical practice, since treatment was determined by physician discretion rather than trial protocol. These results are consistent

with Pivotal Phase 3 Study 2017-01 results, demonstrating positive Treatment Success for patients with rCDI.

Figure 18: RBX2660 Open-Label Studies - Treatment Results Across Studies



7 CLINICAL SAFETY

Summary

- Safety data from the Pivotal Phase 3 Study 2017-01, as well as other individual studies and the integrated safety analyses of data from across the prospective studies of RBX2660, were analyzed.
- The safety profile was consistent across the clinical program.
- Regarding safety in Phase 3 Study 2017-01 during the initial 8 week double-blind period and censoring data at the time of CDI recurrence:
    - AEs were reported for 47.8% (86/180) of patients in the RBX2660 and 39.1% (34/87) of patients in the placebo group, and most AEs in patients treated with RBX2660 were mild or moderate in severity.
        - The most common AEs were gastrointestinal, with the most common being diarrhea (12.2% [22/180] in the RBX2660 group and 12.6% [11/87] in the placebo group) and abdominal pain (12.8% [23/180] in the RBX2660 group and 10.3% [9/87] in the placebo group). The gastrointestinal AEs typically occurred early (within the first 7 days of starting treatment), and were short in duration, lasting a median of 2 days.
    - SAEs were reported for 2.2% (4/180) of patients in the RBX2660 group and 1.1% (1/87) of patients in the placebo group.
    - One death was reported during the double-blind period of Study 2017-01; the death was from cardio-respiratory arrest.
- The integrated safety population included all patients who received at least 1 dose of study treatment in the three Phase 2 studies and two Phase 3 studies through 6 months after treatment. Regarding safety findings in this integrated population:
    - AEs were reported for 68.8% (673/978) of patients in the All RBX2660 group and most AEs were mild or moderate in severity.
    - The most common AEs were gastrointestinal in nature, with the most common being:
        - Diarrhea: 23.1% (226/978)
        - Abdominal pain: 16.4% (160/978)
        - Nausea: 9.3% (91/978)
    - SAEs were reported for 13.8% (135/978) of patients in the All RBX2660 group, with the most common SAE by preferred term being *C. difficile* infection, with a frequency of 2.6% in the All RBX2660 group, and was due to recurrences of CDI meeting SAE criteria. *C. difficile* infection was the only preferred term with >1% frequency, and the remaining SAEs showed no signs of clustering by system organ class.
    - Across all studies, there were 18 AEs with onset within 6 months of last treatment with RBX2660 leading to patient death. There were no deaths among patients given placebo from AEs with onset within 6 months of blinded treatment. However, the exposure time to placebo was 42 patient-years in total, while exposure time for the All RBX2660 group was 404 patient-years. The types of events leading to death were a variety of preferred terms and system organ classes with no pattern or clustering of AEs.
- Overall, RBX2660 was well tolerated, with generally expected and manageable AEs.

7.1 Overview of Safety Program

7.1.1 Safety Data and Analyses

This Briefing Document presents safety data from patients who received one or two doses of blinded treatment in Phase 2B Study 2014-01 or one dose of blinded treatment in the Pivotal

Phase 3 Study 2017-01, as well as integrated safety analyses of data from across the studies in which patients were exposed to RBX2660. A summary of safety results for the Phase 2 Study 2013-001 is also provided in Appendix 11.3.

Safety data were summarized for all patients who received at least 1 study treatment in the three Phase 2 studies and two Phase 3 studies (Table 1 and Table 2). Data from all studies provide safety information and include at least 6 months' follow-up. In addition, data from 2 studies with 24 months of follow-up provide long-term safety data.

In addition, a single retrospective safety and tolerability study (Study 2019-02) was conducted in patients administered RBX2660 for the prevention of rCDI. Given the retrospective study design, this study was excluded from any integrated safety assessments, but safety findings from the study are described in Section 7.6.

7.1.2 Solicited Adverse Events

A patient diary was used in each of the prospective studies to solicit anticipated AEs from patients for the first 7 days after treatment (including after any second assigned study treatment in applicable studies). The diary served as a tool for patients to self-report the incidence and severity of the predefined AEs.

An event reported in the patient diary was not necessarily a reportable AE; rather, the events were reviewed by the investigator to assess the frequency and severity of events and to determine if an AE report was warranted. An AE may have been reported if the investigator's assessment determined that an event occurred at an increased frequency or had worsened in severity after treatment.

The solicited AEs were as follows:

- Gas or flatulence
- Abdominal distension or bloating
- Rectal irritation or pain
- Chills/severe shivering
- Abdominal pain or cramping
- Increased diarrhea
- Constipation
- Rectal bleeding
- Nausea
- Vomiting
- Fever ≥ 38.0°C (in Studies 2013-001, 2014-01, and 2015-01) or 37.8°C (in Studies 2017-01 and 2019-01)

Solicited AEs were analyzed using discrete daily diary data; if more than 1 diary was available for a given day, the record with the highest severity was used in the analysis.

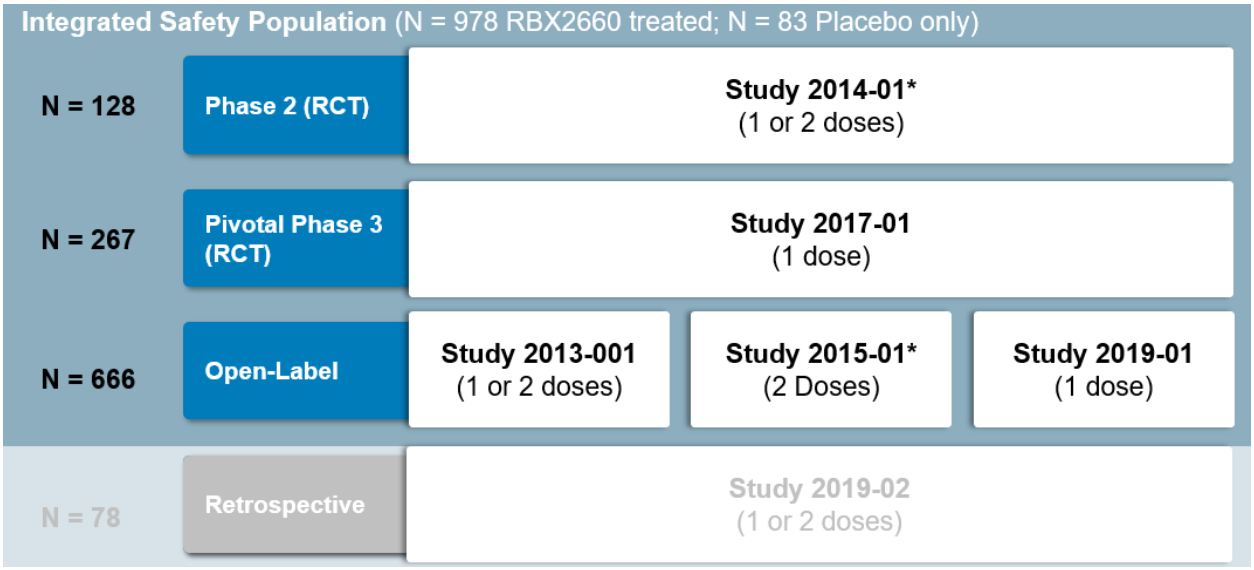
7.1.3 Identification of Adverse Events of Special Interest

Adverse events of special interest (AESIs) identified for safety analyses in accordance with FDA recommendations after trial completion (rather than being reported specifically during the trials) were hyperglycemia/new onset diabetes mellitus and immune-mediated/autoimmune disorders.

7.2 Treatment Exposure

The integrated safety population includes data from placebo-controlled and open-label studies, and also includes patients who received 2 doses of RBX2660 in either a single treatment course or multiple treatment courses. It includes 978 patients treated with RBX2660 and 83 patients who received only placebo (Figure 19). The placebo-controlled data from the Pivotal Phase 3 Study 2017-01, which is the largest study in the program, provides the best assessment of comparative safety data for the selected treatment regimen. Data from the retrospective Study 2019-02 are not included in the integrated safety population.

Figure 19: Safety Exposures



*Included 2 doses of investigational product within each course of treatment.

7.3 Safety in Phase 2B Study 2014-01

Study 2014-01 was a prospective, multicenter, randomized, double-blinded, placebo-controlled, 3-arm Phase 2B study investigating the efficacy and safety of RBX2660 for the treatment of rCDI, and was used to determine the dosing regimen of RBX2660 for subsequent confirmation in the pivotal Phase 3 trial.

The number of AEs was slightly higher in the blinded RBX2660 groups compared to the Blinded Placebo-only group. The rates of moderate and severe AEs were higher in the RBX2660 treatment groups compared to the placebo-only treatment group (Table 10). The rate for SAEs was higher in the RBX2660 treatment groups compared to the placebo-only treatment group. Serious AEs reported during the 8-week double-blind period in Study 2014-01 are summarized by preferred term in Table 11. No SAE preferred MedDRA term was reported in more than one patient, and there were no apparent patterns in the types of reported events. No patients were discontinued due to an AE. There were two AEs leading to death in the group treated with 2 courses of RBX2660 during the 8-week double-blind period (see Section 7.5.6 for additional information on patient deaths).

Data from this study from treatment start through 6 months after completing treatment is also included in the integrated safety analysis.

Table 10: Study 2014-01 8-Week Double-Blind Period – Overview of Adverse Events

	Blinded RBX2660 x 2, n (%) N = 42	Blinded RBX2660 + Placebo, n (%) N = 42	Blinded Placebo x 2, n (%) N = 44
All AEs	26 (61.9%)	29 (69.0%)	25 (56.8%)
Number of AEs	149	80	70
AEs by maximum severity**			
Mild	8 (19.0%)	13 (31.0%)	17 (38.6%)
Moderate	9 (21.4%)	12 (28.6%)	7 (15.9%)
Severe	7 (16.7%)	4 (9.5%)	1 (2.3%)
Potentially life-threatening	2 (4.8%)	0	0
Patient discontinued from study due to AE	0	0	0
All SAEs	8 (19.0%)	5 (11.9%)	1 (2.3%)
Deaths	2 (4.8%)	0	0

Note: Treatment Failures censored at CDI recurrence.
** Severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria; missing severity counted at maximum.

Table 11: Study 2014-01 8-Week Double-Blind Period – Serious Adverse Events by Preferred Term

SAE Preferred Term	Blinded RBX2660 x 2, n (%) N = 42	Blinded RBX2660 + Placebo, n (%) N = 42	Blinded Placebo x 2, n (%) N = 44
Any SAE	8 (19.0%)	5 (11.9%)	1 (2.3%)
Abdominal pain	1 (2.4)	-	-
Abdominal pain upper	-	1 (2.4)	-
Acute respiratory failure	1 (2.4)	-	-
Anemia	-	1 (2.4)	-
Back pain	1 (2.4)	-	-
Constipation	1 (2.4)	-	-
Diabetic neuropathy	-	1 (2.4)	-
Intestinal ischaemia	1 (2.4)	-	-
Intestinal obstruction	-	1 (2.4)	-
Leukocytosis	1 (2.4)		-
Nephrolithiasis	1 (2.4)	-	1 (2.3)
Osteomyelitis chronic		1 (2.4)	-
Renal impairment	1 (2.4)	-	-
Ureteric stenosis	1 (2.4)	-	-
Urinary tract infection	1 (2.4)	-	-

Note: Treatment Failures censored at CDI recurrence.

7.4 Safety in Pivotal Phase 3 Study 2017-01

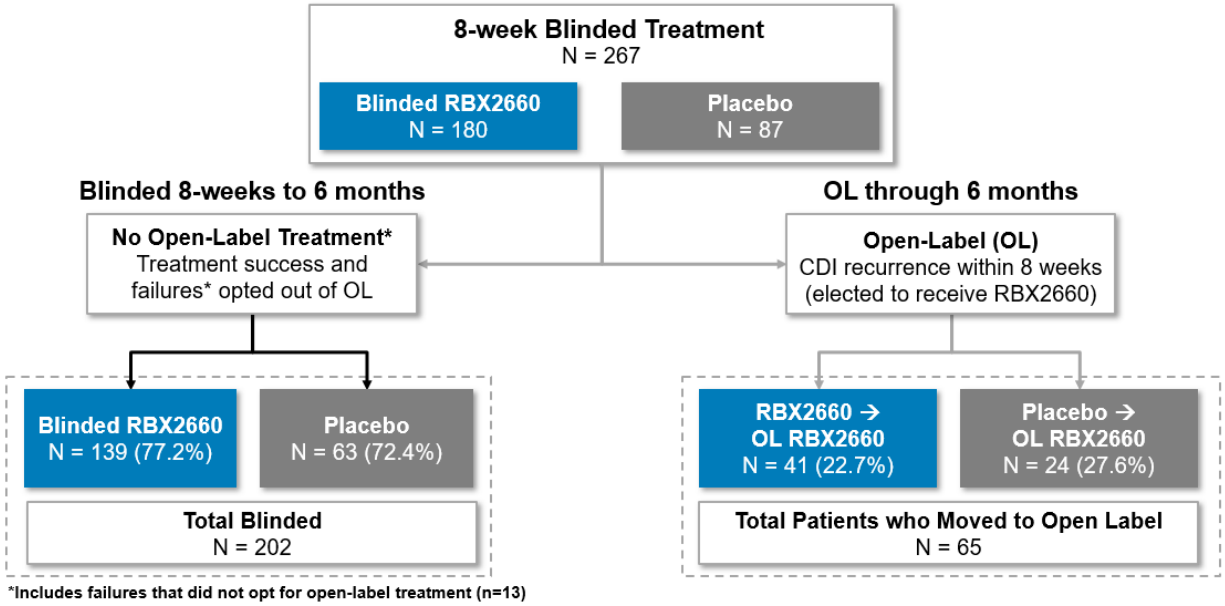
Study 2017-01 was a Phase 3 prospective, multicenter, randomized, double-blind, placebo-controlled study to evaluate the efficacy and safety of RBX2660 for the prevention of rCDI in

patients with documentation of rCDI per the study definition, including either 1) at least 1 recurrence after a primary episode and had completed at least 1 round of standard of care oral antibiotic therapy or 2) had at least 2 episodes of severe CDI resulting in hospitalization within the last year. Patients were randomized to RBX2660 or placebo in a 2:1 ratio.

7.4.1 Study 2017-01: Safety Population Disposition

The safety population for Study 2017-01 consists of 267 patients who were randomized and treated: 180 to RBX2660 and 87 to placebo (Figure 20).

Figure 20: Study 2017-01 Safety Population Disposition



7.4.2 Analysis by Treatment Assignment Censoring at CDI Onset.

In Study 2017-01, the primary efficacy endpoint is collected after 8 weeks. This initial double-blind, placebo-controlled period is followed by a 4-month follow-up period, for a total of 6 months of follow-up. If patients had a Treatment Failure during the initial blinded 8-week period, i.e. experienced a recurrence of CDI within 8 weeks of blinded treatment, they were given the option to enter the open-label part of the trial and receive active RBX2660 treatment followed by 6 months of follow-up. This introduces complexity when reviewing the safety data, because some (N=24/87) of the placebo-treated patients then also become RBX2660-treated patients.

In order to address this complexity, the safety data from the double-blind period for the randomized and treated safety population are presented in this document and include patients from the point after they received active or placebo treatment, to the point at which a patient either completed the specified follow-up time period, ,was lost-to-follow-up**,** or experienced a recurrence of CDI. Patients who experienced a CDI after treatment were censored patients at the day of CDI recurrence.

This analysis allows the best comparison of placebo-controlled safety data, since it removes many confounding safety variables. The primary endpoint of this trial, recurrence of CDI, is a significant medical event that is often causative of sequelae such as diarrhea, abdominal pain, hospitalization, and antibiotic treatment, which may imbalance the safety analyses.

7.4.2.1 Study 2017-01 Double-Blind Period: Overview of Adverse Events

Adverse events discussed in this document are ones that were treatment-emergent (ie, the onset occurred on or after the initial study treatment date).

Rates of AEs during the 8-Week Double-Blind Period were higher in the RBX2660 group compared to placebo and were mostly driven by a higher rate of mild events (Table 12). The rates of moderate and severe AEs were balanced between the treatment groups. The rate for SAEs was also comparable between the treatment groups. No patients were discontinued due to an AE or SAE. There was one death in the RBX2660 group.

Table 12: Study 2017-01 8-Week Double-Blind Period – Overview of Adverse Events

	Blinded RBX2660 N = 180	Placebo N = 87
All AEs	86 (47.8%)	34 (39.1%)
Number of AEs	262	87
AEs by maximum severity*		
Mild	40 (22.2%)	13 (14.9%)
Moderate	37 (20.6%)	18 (20.7%)
Severe	8 (4.4%)	3 (3.4%)
Potentially life-threatening**	1 (0.6%)	0
Patient discontinued from study due to AE**	1 (0.6%)	0
All SAEs	4 (2.2%)	1 (1.1%)
Deaths**	1 (0.6%)	0

Note: Treatment failures are censored at time of CDI recurrence.
* AEs reported by maximum severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria.
** Same patient represented in each category.

7.4.2.2 Study 2017-01 Double-Blind Period: Common Adverse Events

The most commonly reported AEs (reported by ≥ 5% of all patients) in Study 2017-01 during the 8-Week Double-Blind Period are summarized in Table 13. The most common AEs (occurring in ≥ 5% of patients) were gastrointestinal in nature, which is generally expected in treating this patient population; these AEs typically occurred within the first 7 days after treatment and lasted for a median of 2 days. An approximately equal proportion of patients experienced diarrhea in either treatment group, and a greater proportion of patients treated with RBX2660 reported abdominal pain or nausea compared to patients receiving placebo.

Table 13: Study 2017-01 8-Week Double-Blind Period – Adverse Events Reported in ≥ 5% of Patients

	Blinded RBX2660 N = 180	Placebo N = 87
Patients with AEs	86 (47.8%)	34 (39.1%)
Gastrointestinal disorder	53 (29.4%)	26 (29.9%)
Diarrhea	22 (12.2%)	11 (12.6%)
Abdominal pain	23 (12.8%)	9 (10.3%)
Nausea	14 (7.8%)	4 (4.6%)

Note: Treatment failures are censored at time of CDI recurrence.

7.4.2.3 Study 2017-01 Double-Blind Period: Serious Adverse Events

Overall, few patients experienced SAEs in either treatment arm during the 8-Week Double-Blind Period (Table 14). Four patients treated with RBX2660 experienced 6 SAEs, all of which were single events with no common etiology, and one patient in the placebo group reported an SAE. The SAEs generally resolved without sequalae, except for the event of asthenia, which occurred in a patient with a medical history of asthenia; the investigator reported this event as resolving at study exit.

Table 14: Study 2017-01 8-Week Double-Blind Period – Serious Adverse Events

Preferred Term		Blinded RBX2660 N = 180	Placebo N = 87
Patients with SAEs		4 (2.2%)	1 (1.1%)
Patient #1	Ileus	1 (0.6%)	0
	Abdominal abscess	1 (0.6%)	0
Patient #2	Asthenia	1 (0.6%)	0
	Hand fracture	1 (0.6%)	0
Patient #3	Abdominal Pain	1 (0.6%)	0
Patient #4	Cardio-respiratory arrest*	1 (0.6%)	0
Patient #5	Cellulitis	0	1 (1.1%)

*SAE that led to discontinuation and death; see Section 7.4.4 for additional details.

7.4.2.4 Study 2017-01 Overview of Adverse Events Through 6 Months

Safety results through 6 months after blinded treatment were generally similar to those for the 8-Week Double-Blind Period. A higher rate of AEs was reported in the RBX2660 group (55.6%; 100/180) compared to the Placebo group (44.8%; 39/87), which was driven primarily by patients experiencing a mild event by maximum severity (Table 15). Serious AEs were reported for 3.9% (7/180) of blinded RBX2660 patients through 6 months, compared with 2.3% (2/87) in the blinded Placebo group. The same patient who experienced an AE leading to death in the first 8 weeks of double blind treatment remained the only patient who died through 6 months in this analysis

Table 15: Study 2017-01 Safety Overview Through 6 Months

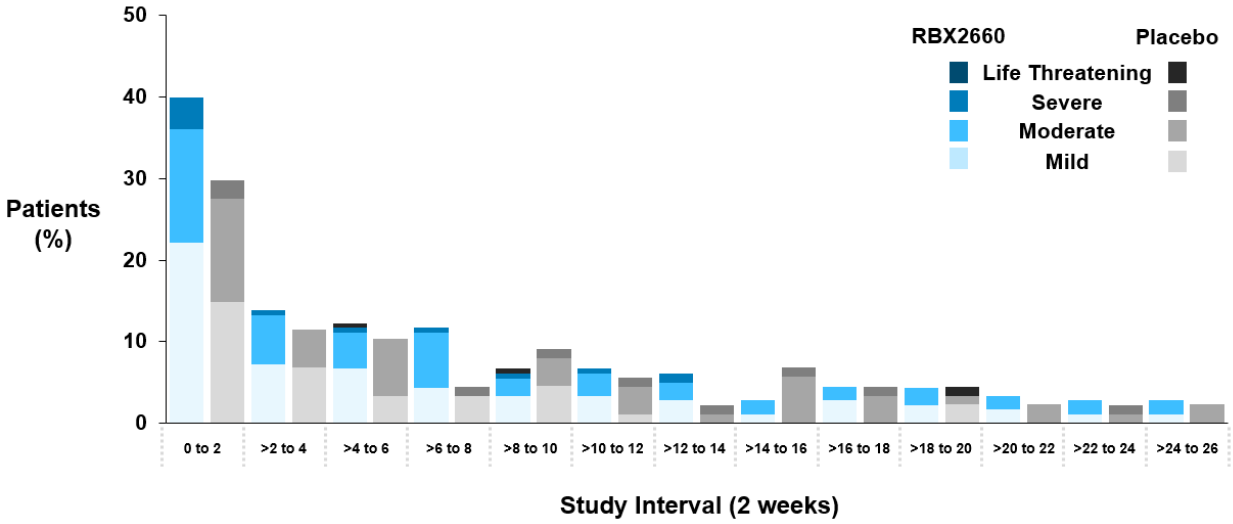
	Blinded RBX2660 N = 180	Blinded Placebo N = 87
AEs	100 (55.6%)	39 (44.8%)
Number of AEs	368	131
AEs by maximum severity*		
Mild	42 (23.3%)	9 (10.3%)
Moderate	47 (26.1%)	25 (28.7%)
Severe	10 (5.6%)	5 (5.7%)
Potentially life-threatening**	1 (0.6%)	0 (0.0)
Patient discontinued from study due to AE**	1 (0.6%)	0 (0.0)
SAEs	7 (3.9%)	2 (2.3%)
Deaths**	1 (0.6%)	0 (0.0)

Note: Treatment failures are censored at time of CDI recurrence.
* AEs reported by maximum severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria.
** Same patient represented in each category.

In addition, an analysis of the onset interval of AEs after blinded treatment through 6 months, further delineated by the maximum severity of AEs, was performed (Figure 21). Most AEs

occurred during the first 2 weeks on treatment, were predominantly mild to moderate in both groups, and the difference between RBX2660 and Placebo was primarily attributable to patients experiencing mild events by maximum severity. After the initial 2-week interval, the proportion of patients with AEs declined in subsequent 2-week intervals, with comparable rates of AEs between patients receiving RBX2660 and placebo.

Figure 21: Study 2017-01 Adverse Events by Severity Through 6 Months



7.4.3 Safety by Treatment Received

In this analysis of safety, patients were grouped according to whether they received one or two courses of treatment. As previously described, patients who had a confirmed recurrence of CDI within 8 weeks after administration of the last assigned study dose were considered Treatment Failures and were offered an open-label treatment course of RBX2660. Regardless of treatment outcome, patients were monitored through 6 months after receiving the last dose of RBX2660, as defined in the study protocol.

7.4.3.1 Study 2017-01 Safety Overview from Treatment to 8 Weeks

Table 16 presents an overview of the safety data from Study 2017-01 through 8 weeks of treatment. In patients who received only one course of blinded treatment, there were more patients who experienced AEs in the RBX2660-only group than in the Placebo-only group. This difference was attributable to patients experiencing mild AEs (by maximum severity). For patients who received two courses of treatment, their safety data is presented separately by AEs experienced after the first course of treatment versus AEs experienced after the second course of treatment (which was Open-label RBX2660). Note that these patients must have experienced a recurrence of CDI during the blinded treatment period to be eligible for open-label RBX2660 treatment. In the blinded portion of the study for patients who received two treatment courses, patients who received blinded placebo or RBX2660 had comparable rates of AEs. The lower rates of AEs during the blinded period in patients in both the RBX2660 and placebo-treated groups who later received a second course or treatment, compared to the patients who only received a single course of blinded therapy, may reflect their shorter duration of observation because their 8-week follow-up was interrupted by recurrence of CDI.

In the open-label period, the rates of AEs in the patients who received open-label treatment were consistent with the groups of patients who only received one treatment course. The rates of patients experiencing AEs in those who received only blinded RBX2660 and those that received open label RBX2660 were similar through 8 weeks of follow-up after their last treatment (56.8% vs 56.1%). Between these two groups, there were some differences in the percentage of patients with events of mild to moderate severity (a decrease in mild events but an increase in moderate events), with a higher rate of discontinuations due to AEs and of SAEs. Despite these numerical differences, the data support that RBX2660 can be safely administered a second time, if a patient experiences a CDI recurrence after the first administration.

Table 16: Study 2017-01 - Safety Overview from Treatment to 8 Weeks by Treatment Received

	Blinded Period (First Treatment)				Open-Label Period (Second treatment)	
From Treatment to 8 weeks	**Blinded RBX2660 N = 139**	**Placebo Only N = 63**	**Blinded RBX2660/ OL RBX2660 N = 41**	**Placebo/ OL RBX2660 N=24**	**Blinded RBX2660/ OL RBX2660 N = 41**	**Placebo/ OL RBX2660 N=24**
All AEs	79 (56.8)	30 (47.6)	15 (36.6)	8 (33.3)	23 (56.1%)	11 (45.8)
Number of AEs	258	84	42	17	48	17
AEs by maximum severity*						
Mild	36 (25.9)	8 (12.7)	4 (9.8)	4 (16.7)	8 (19.5%)	5 (20.8)
Moderate	32(23.0)	17 (27.0)	8 (19.5)	4 (16.7)	12 (29.3%)	6 (25.0)
Severe	10 (7.2)	5 (7.9)	3 (7.3)	0 (0.0)	3 (7.3%)	0 (0.0)
Potentially life-threatening**	1 (0.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)
AE Leading to Discontinuation**	1 (0.7)	0 (0.0)	0 (0.0)	0 (0.0)	1 (2.4%)	0 (0.0)
All SAEs	6 (4.3)	3 (4.8)	3 (7.3)	1 (4.2)	3 (7.3%)	1 (4.2)
Deaths**	1 (0.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)

Note: The table shows all AEs within the first 8 weeks, i.e., Treatment Failures are *not* censored at time of CDI recurrence.
* AEs reported by maximum severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria.
** Same patient represented in each category.

** AEs leading to discontinuation include AEs Leading to Death.

7.4.3.2 Study 2017-01 Safety: Safety Overview Through 6 Months After Open-Label RBX2660 Treatment

Safety findings in the open-label portion of Study 2017-01 through 6 months after open-label RBX2660 treatment are summarized in Table 17. Overall, there were comparable event rates between the treatment groups, and AEs are less frequent than during the first 8 weeks after treatment (Table 16). Adverse events leading to discontinuation were reported by 4.9% of patients in the RBX2660-randomized patients who went on to receive open-label RBX2660, compared to no AEs leading to discontinuation reported in placebo-randomized patients who went on to receive open-label RBX2660.

One death occurred in the blinded RBX2660/open-label RBX2660 group during this period; additional details on deaths that occurred in Study 2017-01 are provided in Section 7.4.4.

Table 17: Study 2017-01 Open-Label Portion – Safety Overview Through 6 Months After Open-Label RBX2660 Treatment

Through 6-months after Open Label treatment	**Blinded RBX2660/ Open-Label RBX2660 N = 41**	**Placebo/ Open-Label RBX2660 N = 24**
All AEs	24 (58.5%)	14 (58.3%)
Number of AEs	89	43
AEs by maximum severity*		
Mild	8 (19.5%)	6 (25.0%)
Moderate	10 (24.4%)	6 (25.0%)
Severe	5 (12.2%)	1 (4.2%)
Potentially life-threatening	1 (2.4%)	1 (4.2%)
Patient discontinued from study due to AE	2 (4.9%)	0
All SAEs	5 (12.2%)	1 (4.2%)
Deaths	1 (2.4%)	0

* AEs reported by maximum severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria.

7.4.4 Study 2017-01: Deaths

Two patients died during the entirety of Study 2017-01 (Table 18). The first patient was a 75-year-old male who died of cardio-respiratory arrest 37 days after RBX2660 treatment (within the double-blind period); he suffered from several comorbid conditions, including multiple cardiovascular and central nervous system diseases. This patient's death was reported as unrelated to study treatment, as determined by both the Investigator and the Data Safety Monitoring Board (DSMB).

The second patient who died was a 79-year-old female with a history of cardiac disease, diabetes, CNS disease, and chronic kidney disease. She died 151 days after the last treatment with RBX2660; the cause of death was due to multimorbidity and was assessed as unrelated to study treatment by both the Investigator and the DSMB.

Table 18: Study 2017-01: Deaths During the Study

Age/ Sex	Treatment Details	Cause of Death	Day of Death from last RBX2660 treatment	Select Medical History/ Key Comorbidities	Related to Study Drug (Y/N)*
75 yrs Male	Blinded RBX2660	Asystole, cardio-respiratory arrest	Day 37	Coronary artery bypass grafting (CABG) × 4, mixed hyperlipidemia, atrial fibrillation, tachycardia, hypertension, cerebrovascular accident, epilepsy, anemia, Parkinson's disease, gastroesophageal reflux disease	N
79 yrs Female	Blinded RBX2660 + Open-label RBX2660 at Week 4	Multimorbidity	Day 151	Recurrent CDI, congestive heart failure, hypertension, hyperlipidemia, Diabetes Mellites Type 2, diabetic neuropathy, chronic kidney disease, depression, anxiety, anemia, chronic urinary tract infections	N

*Relatedness determined by Investigator, and reviewed by DSMB.

7.4.5 Study 2017-01: Solicited Adverse Events

During the double-blind period, 94.4% (170/180) of patients from the RBX2660 group and 96.6% (84/87) of patients in the placebo group experienced at least one solicited AE. During the open-label period, 87.7% (57/65) of patients experienced at least one solicited AE.

The majority of solicited AEs reported by patients were assessed as mild or moderate. The solicited events reported as severe by patients via patient diary included, in decreasing order of frequency, abdominal pain or cramping (33 patients, 12.4%), increased diarrhea (30 patients, 11.2%), abdominal distension or bloating (22 patients, 8.2%), rectal irritation or pain (10 patients, 3.7%), nausea (9 patients, 3.4%), chills/severe shivering (5 patients, 1.9%), constipation (4 patients, 1.5%), fever (2 patients, 0.7%), vomiting (2 patients, 0.7%), rectal bleeding (1 patient, 0.4%).

7.5 Safety Across All Studies

7.5.1 Integrated Safety Population and Disposition

The integrated safety population included 978 patients in the RBX2660 group (Table 19), the majority of whom were from open-label studies. The majority of patients treated with placebo came from Study 2017-01.

Baseline demographics and characteristics for patients in the integrated safety population are summarized Table 31 in Appendix 11.4.

Disposition of the integrated safety population is summarized in Table 20; the 4 defined groups (other than the All RBX2660 group) are mutually exclusive (ie, each patient belongs to one group only).

Patients who received only 1 treatment course are included in the Placebo Only or RBX2660 Only groups. If a patient received a second course of open-label RBX2660 treatment following the recurrence of CDI, they are separated depending on their first course of treatment. The number of protocol-defined exposures to RBX2660 ranged from 1 to 4 doses. The Phase 3 studies used a dosing regimen of 1 dose per treatment course. If a patient experienced a recurrence of CDI after the first treatment course, a second treatment course was offered, again consisting of 1 dose.

However, in Studies 2014-01 and 2015-01, 1 treatment course consisted of 2 doses administered 7 ± 2 days apart. In Study 2014-01, as the first treatment course, patients were randomized to receive 1) 2 doses of RBX2660, 2) 2 doses of placebo, or 3) 1 dose of RBX2660 and 1 dose of placebo (Table 1). Patients could have received an additional treatment course consisting of up to 2 doses of RBX2660 if they had a confirmed recurrence within 8 weeks of the first course. Patients in Study 2014-01 could have had up to 4 RBX2660 exposures.

Some included data are derived from the ongoing open-label Study 2019-01 (data cutoff date: 25 Mar 2022), which is examining safety.

All clinical studies of RBX2660 included at least 6 months of safety follow-up after the last dose of RBX2660. Studies 2014-01 and 2015-01 included a follow-up duration of 24 months after the last dose of RBX2660. To standardize the duration of follow-up across all studies, the primary safety analyses were from baseline first exposure through 6 months after completing treatment. If a patient received a second treatment course, the follow-up day was reset to day 1, so that patients were followed for 6-months after their last treatment.

Table 19: Integrated Safety Populations by Treatment Group

	Placebo Only (N = 83)	RBX2660 Only (N = 763)	Placebo / Open-Label RBX2660 (N = 48)	RBX2660 / Open-Label RBX2660 (N = 167)	All RBX2660 (N = 978)
	One treatment course*		Two treatment courses*		
Study 2013-001 (Open-label)	0 (0.0)	19 (2.5%)	0 (0.0)	15 (9.0%)	34 (3.5%)
Study 2014-01 (DB, PBO Study)	20 (24.1%)	54 (7.1%)	24 (50.0%)	30 (18.0%)	108 (11.0%)
Study 2015-01 (Open-label)**	0 (0.0)	149 (19.5%)	0 (0.0)	0 (0.0)	149 (15.2%)
Study 2017-01 (DB, PBO Study)	63 (75.9%)	139 (18.2%)	24 (50.0%)	41 (24.6%)	204 (20.9%)
Study 2019-01 (Open-label)	0 (0.0)	402 (52.7%)	0 (0.0)	81 (48.5%)	483 (49.4%)

*One treatment course can be one or two doses
** Study 2015-01 did not have an option for a second course of treatment.

Table 20: Integrated Safety Population Disposition

	Placebo Only (N = 83)	RBX2660 Only (N = 763)	Placebo / Open-Label RBX2660 (N = 48)	RBX2660 / Open-Label RBX2660 (N = 167)	All RBX2660 (N = 978)
	One treatment course*		Two treatment courses*		
Received treatment	83 (100.0%)	763 (100.0%)	48 (100.0%)	167 (100.0%)	978 (100.0%)
Completed 8-week follow-up (after first treatment)	78 (94.0%)	672 (88.1%)	42 (87.5%)	146 (87.4%)	860 (87.9%)
Completed 6-month follow-up (after first treatment)	75 (90.4%)	583 (76.4%)	42 (87.5%)	126 (75.4%)	751 (76.8%)
Completed 24-month follow-up (after first treatment)**	16 (19.3%)	145 (19.0%)	19 (39.6%)	18 (10.8%)	182 (18.6%)
Ongoing in 2019-01***	0 (0.0)	119 (15.6%)	0 (0.0)	25 (15.0%)	144 (14.7%)
Between treatment and 8-week follow-up	0 (0.0)	38 (31.9%)	0 (0.0)	6 (24.0%)	44 (30.6%)
Between 8-week and 6-	0 (0.0)	68 (57.1%)	0 (0.0)	16 (64.0%)	84 (58.3%)

*One treatment course can be one or two doses
**Only Study 2014-01 and Study 2015-01 included a follow-up duration of 24 months.

*7.5.2 *** Data cutoff date for study 2019-01 is 25Mar2022. Patients may have had 6-months of follow-up but not completed Study Exit form, thus considered "ongoing." Integrated Safety Population: Overview of Adverse Events in Blinded Studies*

To assess the integrated safety results from blinded studies, safety data from patients in Studies 2014-01 and 2017-01 who received only blinded RBX2660 or Placebo was pooled. This analysis excluded patients who received open-label RBX2660 after initial treatment failure. A higher rate of AEs in patients receiving blinded RBX2660 versus Placebo was observed, which was attributable to patients experiencing a mild AE by maximum severity (Table 21).

Table 21: Integrated Safety Population - Overview of Adverse Events in Blinded Studies

Safety through 6-months after last	Blinded RBX2660 Only (1 or 2 doses) N = 193	Placebo Only N = 83
All patients with AEs	135 (69.9%)	50 (60.2%)
Number of AEs	632	174
AEs by maximum severity*		
Mild	56 (29.0%)	13 (15.7%)
Moderate	54 (28.0%)	29 (34.9%)
Severe	19 (9.8%)	7 (8.4%)
Potentially life-threatening	6 (3.1%)	1 (1.2%)
Patient discontinued from study due to AE**	1 (0.5%)	0 (0.0)
All SAEs	20 (10.4%)	6 (7.2%)
Deaths	5 (2.6%)	0 (0.0)

* AEs reported by maximum severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria;
** AEs leading to discontinuation were only collected in Studies 2017-01 and 2019-01, which also includes deaths in these studies.

7.5.3 Integrated Safety Population: Overview of Adverse Events

For the integrated safety population across all five prospective studies, 68.8% of the patients treated with RBX2660 experienced AEs (Table 22). The severity of most AEs was mild or moderate. Potentially life-threatening AEs were reported infrequently, being experienced by 3.0% of patients, and SAEs were reported for 13.8% of patients treated with RBX2660. As aforementioned, the All RBX2660 group represents all patients who were exposed to RBX2660, and includes patients who received one or two courses of treatment, including patients who received blinded Placebo, and then received open-label RBX2660 whereas the Placebo Only represents the potentially healthier population who did not have a recurrence of rCDI. A direct comparison should be made with caution due to different numbers of treatment courses as well as doses in the different groups and the follow-up time. Of note, placebo was only used in the two randomized trials, whereas active treatment was included in all trials. This bias should be taken into consideration when reviewing the safety table below,

Patients in the RBX2660 1 Dose Only group were assigned to receive one dose of RBX2660 only, which may have occurred in controlled or open-label studies. Results for the RBX2660 1 Dose Only and Placebo Only groups were generally similar to the All RBX2660 group, except for SAEs and deaths in the Placebo Only group as would be expected based on the biases described above.

Table 22: Integrated Safety Population – Overview of Adverse Events in All Patients Treated with RBX2660

Safety through 6-months after last treatment*	All RBX2660 (1-4 Doses) N = 978	RBX2660 1 Dose Only N = 595	Placebo Only N = 83
All patients with AEs	673 (68.8%)	378 (63.5%)	50 (60.2%)
Number of AEs	2881	1327	174
AEs by maximum severity**			
Mild	224 (22.9%)	124 (20.8%)	13 (15.7%)
Moderate	294 (30.1%)	176 (29.6%)	29 (34.9%)
Severe	126 (12.9%)	64 (10.8%)	7 (8.4%)
Potentially life-threatening	29 (3.0%)	14 (2.4%)	1 (1.2%)
Patient discontinued from study due to AE***	8 (0.8%)	5 (0.8%)	0 (0.0)
All SAEs	135 (13.8%)	60 (10.1%)	6 (7.2%)
Deaths	18 (1.8%)	5 (0.8%)	0 (0.0)

Note: Treatment failures are censored at time of CDI recurrence.
* AEs after the first treatment in patients who received a second treatment are included; the All RBX2660 group includes those with Treatment Failure on placebo who crossed over to RBX2660 treatment.
** AEs reported by maximum severity as assessed by investigator using Common Terminology Criteria for Adverse Events (CTCAE) criteria;
*** AEs leading to discontinuation were only collected in Studies 2017-01 and 2019-01, which also includes deaths in these studies.

7.5.4 Integrated Safety Population: Common Adverse Events

Across the treatment groups, gastrointestinal disorders were the most frequently reported AEs by System Organ Class. Regarding AEs reported in ≥ 5% of patients treated with RBX2660, diarrhea and abdominal pain were the most frequently reported (Table 23).

Table 23: Integrated Safety Population – Adverse Events Reported in ≥ 5% of Patients Treated with RBX2660

Preferred Term	All RBX2660 (1-4 Doses) (N = 978)	RBX2660 1 Dose Only N = 595	Placebo Only N = 83
Any AE	673 (68.8%)	378 (63.5%)	50 (60.2%)
Diarrhea	226 (23.1%)	113 (19.0%)	15 (18.1%)
Abdominal pain	160 (16.4%)	90 (15.1%)	7 (8.4%)
Nausea	91 (9.3%)	54 (9.1%)	3 (3.6%)
Flatulence	72 (7.4%)	43 (7.2%)	1 (1.2%)
Abdominal distension	69 (7.1%)	39 (6.6%)	3 (3.6%)
Urinary tract infection	64 (6.5%)	27 (4.5%)	4 (4.8%)
Constipation	63 (6.4%)	22 (3.7%)	5 (6.0%)
Upper respiratory tract infection	23 (2.4%)	12 (2.0%)	5 (6.0%)

7.5.5 Integrated Safety Population: Serious Adverse Events

SAEs were reported in 13.8% of the patients treated with RBX2660 (Table 24). Infections and infestations were the most common types of SAEs by System Organ Class (5.6% of patients), with 2.6% of patients reporting SAEs of *C. difficile* infection by preferred term (note that *C. difficile* infection was only an SAE when a recurrence happened and met SAE criteria).

Table 24: Integrated Safety Population – Serious Adverse Events in ≥ 1% of All RBX2660-Treated Patients

System Organ Class	All RBX2660 (1-4 Doses) (N = 978)	RBX2660 1 Dose Only N = 595	Placebo Only N = 83
Any SAE*	135 (13.8%)	60 (10.1%)	6 (7.2%)
Cardiac disorders	16 (1.6%)	6 (1.0%)	0 (0.0)
Gastrointestinal disorders	29 (3.0%)	9 (1.5%)	1 (1.2%)
General disorders and administration site conditions	11 (1.1%)	5 (0.8%)	0 (0.0)
Infections and infestations*	55 (5.6%)	30 (5.0%)	4 (4.8%)
Injury, poisoning and procedural complications	15 (1.5%)	6 (1.0%)	0 (0.0)
Metabolism and nutrition disorders	10 (1.0%)	4 (0.7%)	1 (1.2%)
Renal and urinary disorders	10 (1.0%)	3 (0.5%)	1 (1.2%)
Respiratory, thoracic and mediastinal disorders	20 (2.0%)	8 (1.3%)	1 (1.2%)

* 2.6% Frequency of CDI as an SAE driven by CDI recurrences requiring hospitalization.

7.5.6 Integrated Safety Population: Deaths

Regarding patient deaths across all studies, there were 18 deaths due to AEs with an onset within 6 months after the last treatment course, all of which occurred in the RBX2660 group (Table 25). the majority (11/18) of the patients with AEs leading to death had onset of the AE after the 8-week efficacy timepoint. (Patient deaths during the subsequent long-term follow-up are discussed in Section 7.7.)

There were two patient deaths within 30 days of the last dose of RBX2660:

- One patient experienced a severe recurrence of CDI 14 days after the second (and final) dose of RBX2660 in study 2015-01 and died 10 days later. This patient was a 94-year old female, who suffered from chronic kidney disease (stage IV), hypertension, hyperglycemia, gastroesophageal reflux disease, depression with anxiety, anemia in chronic disease, and rCDI. The cause of death was *C. difficile* infection. The fatal event was assessed by the investigator as definitely related to pre-existing conditions and CDI and possibly related to RBX2660 and the enema procedure. The product the patient received was tested and found negative for *C. difficile*. This event was subsequent reviewed by an Independent Medical Monitor who deemed that the event did not constitute a product safety concern.
- The second patient death involved a patient in Study 2014-01 who experienced a recurrence of CDI after receiving one blinded dose of RBX2660 and one dose of Placebo. The patient elected to be re-treated with Open-Label RBX2660 and received two additional doses. Twenty -five days after final dose, the patient experienced bacteremia, sepsis, staphylococcal infection, and respiratory failure. The patient had a history of end stage renal disease, diabetes, foot ulcer, coronary artery disease, heart failure, and a lacunar infarct. Since the patient was undergoing dialysis when symptoms of hypotension and short of breath occurred, the suspected source of the pathogen (blood culture positive for methicillin resistant *S. aureus*) was the dialysis catheter or possibly decubitus ulcers. The patient received multiple antibiotics and surgery, and did not respond to vasopressor support. The patient was placed on palliative care, and died 4 days after the diagnosis of sepsis, a total of 29 days after final RBX2660 dose.

There were 3 patients in Studies 2014-01 and 2015-01, who had onset of an AE leading to death within 6 months of treatment, and resulted in death after the 6-month timepoint. This 6-month timeframe was established to create a consistent time period of follow-up across all studies for the purpose of integrating the safety database, and for the events in these studies which transited the cutoff period, they were included according to the onset date of the event.

As the integrated safety database contains both open-label and placebo-controlled studies, there is a significant imbalance with respect to the number of patients at risk of an AE in the RBX2660 group compared to Placebo. Additionally, because Placebo Treatment Failures were able to be retreated with RBX2660, there is an imbalance in the time patients were at risk of an AE leading to death in RBX2660 vs Placebo. The patients who received only placebo had an overall observational time of 42 patient years, while the RBX2660 treatment group had an observational time of 404 years. Given a combined total of 446 patient years of observation across the RBX2660 and placebo treatment groups, the observation time for the RBX2660 group was approximately nine times (404/446; 90.6%) that of the placebo group.

Please also note that because patients who failed placebo were offered to be treated with RBX2660, the patients who only received Placebo tend to be Treatment Successes.

Table 25: Integrated Safety Population – Adverse Events Leading to Patient Death by Observation Time

	All RBX2660 (N = 978)	Placebo Only (N = 83)
AEs Occurring within 6 months Leading to Death	18 (1.8%)	0
Observation time (patient-years)*	404	42
Date of death (after last treatment)		
1 – 30 days	2	0
>30 – 60 days	5	0
>60 days – 6 months	8	0
>6 months – 2 years	3	0

* Observation time was restricted to 6-months after starting last treatment. The observation time for patients who were assigned to placebo but received RBX2660 after blinded failure is included under Placebo only.

There was no clear pattern in the AEs leading to death for events starting with 6 months of last study treatment, with no apparent clustering of pathologies or preferred terms (Table 26). By-patient narratives for all of these AEs leading to death are provided in Appendix 11.5.

Table 26: Integrated Safety Population – Adverse Events Starting Within 6 Months of Last RBX2660 Leading to Patient Death

System Organ Class/Preferred Term	All RBX2660 (N = 978)
Any AE leading to death with a start date within 6 months of last treatment with RBX2660	18 (1.8)%
Cardiac disorders	**3 (0.3%)**
Cardiac arrest	1 (0.1%)
Cardiac failure congestive	1 (0.1%)
Cardio-respiratory arrest	1 (0.1%)
Congenital, familial and genetic disorders	**1 (0.1%)**
Spina bifida	1 (0.1%)
Gastrointestinal disorders	**1 (0.1%)**
Intestinal ischaemia	1 (0.1%)
General disorders and administration site conditions	**4 (0.4%)**
General physical health deterioration	2 (0.2%)
Death	1 (0.1%)
Multimorbidity	1 (0.1%)
Infections and infestations	**4 (0.4%)**
Sepsis	2 (0.2%)
Bacteraemia	1 (0.1%)
Clostridium difficile infection	1 (0.1%)
Pulmonary sepsis	1 (0.1%)
Staphylococcal infection	1 (0.1%)
Injury, poisoning and procedural complications	**1 (0.1%)**
Pelvic fracture	1 (0.1%)
Renal and urinary disorders	**2 (0.2%)**
Nephropathy	1 (0.1%)
Renal failure	1 (0.1%)
Respiratory, thoracic and mediastinal disorders	**4 (0.4%)**
Respiratory failure	2 (0.2%)
Acute respiratory failure	1 (0.1%)
Chronic obstructive pulmonary disease	1 (0.1%)

7.5.7 Integrated Safety Population: Adverse Events of Special Interest

Based on guidance from the FDA (see Section 7.1.3 for additional details), AESIs were examined, including an analysis of patient data for metabolic and autoimmune disorders. The two SMQs summarized below, "hyperglycemia/new onset diabetes mellitus" and "immune-mediated autoimmune disorders," capture sets of preferred terms of interest that were reported across the clinical program. Overall, few AESIs were reported and were balanced between the treatment groups; hyperglycemia/new onset diabetes mellitus occurred in 1.6% (12/763) of RBX2660 Only patients and 2.4% (2/83) Placebo Only patients, while immune-mediated/autoimmune disorders occurred in 1.3% (10/763) of RBX2660 Only patients and 1.2% (1/83) Placebo Only patients.

7.6 Safety in Retrospective Study 2019-02

The retrospective safety and tolerability Study 2019-02 was conducted in patients administered RBX2660 for the prevention of rCDI. The objective of the study was to evaluate the safety and tolerability of RBX2660 through 6 months after treatment in patients who received RBX2660 and were treated under the discretion of the Investigator without prespecified exclusion criteria. Safety results from the study were generally similar to those for patients treated with RBX2660 in the integrated safety population (Table 27). Of the 40 patients who experienced AEs, the majority experienced a mild or moderate AEs, while 5 of 40 experienced severe or life-threatening AEs by maximum severity. The severe and life-threatening events spanned 5 different system organ classes and were considered unrelated to RBX2660.

Table 27: Retrospective Study 2019-02 – Overview of Adverse Events

	RBX2660 N = 64	
Adverse Event Category	**Events**	**N (%)**
Patients with any AE	144	40 (62.5)
Related AEs		
Related to RBX2660	32	11 (17.2)
Related to enema procedure	4	3 (4.7)
Severe or life-threatening AEs	10	5 (7.8)
AEs leading to death	1	1 (1.6)
Serious AEs	11	8 (12.5)

7.7 Long-Term Safety

Data regarding AEs during long-term follow-up (6 to 24 Months) are available from patients who participated in Studies 2014-01 and 2015-01, and only patients within those studies who did not discontinue before reaching the 6-month timepoint are considered for the following analyses. Overall, a higher percentage of patients in the RBX2660 Only (56.3%) than in the Placebo Only group (47.4%) had AEs between 6 and 24 months after first dose administration (Table 28). Most AEs were mild or moderate in severity. Severe AEs were reported in 7.4% (13 of 176) of patients in the RBX2660 Only group and 10.5% (2 of 19) of patients in the Placebo Only group. A lower percentage of patients in the RBX2660 Only group (7.4%) than in the Placebo Only group (10.5%) had AEs leading to death for events with an onset greater than 6 months after the most recent study treatment.

Table 28: Integrated Safety Population – Overview of Adverse Events From 6 to 24 Months After Administration of First Enema of Most Recent Treatment Course

	RBX2660 Only N = 176	Placebo Only N = 19	RBX2660 / Open-Label RBX2660 N = 23	Placebo / Open-Label RBX2660 N = 23
All patient with AEs	99 (56.3%)	9 (47.4%)	11 (47.8%)	15 (65.2%)
Number of AEs	424	54	24	56
AEs by maximum severity				
Mild	27 (15.3%)	2 (10.5%)	2 (8.7%)	4 (17.4%)
Moderate	36 (20.5%)	3 (15.8%)	3 (13.0%)	4 (17.4%)
Severe	13 (7.4%)	2 (10.5%)	4 (17.4%)	6 (26.1%)
Potentially life-threatening	23 (13.1%)	2 (10.5%)	2 (8.7%)	1 (4.3 %)
AEs Leading to Death	13 (7.4%)	2 (10.5%)	3 (13.0%)	0

8 POST-MARKETING PLAN

The safety profile of RBX2660 summarized in this document did not identify any unexpected adverse reactions across a diverse rCDI patient population, including older adults, immunocompromised patients, and severely ill patients. The important potential risk of the transmission of infection identified for RBX2660 was not observed in clinical trials, but is a possible risk. In addition to the ongoing donor screening and testing requirements, the safety of RBX2660 will be continuously monitored after marketing approval through the pharmacovigilance program.

A group of healthcare professionals, trained to identify AEs will be available by phone to field calls and capture any events. A dedicated call center will also be established to assist patients and healthcare professionals with access to RBX2660 and these representatives will also be trained to identify and report AEs. All AEs are entered into a global safety database and Periodic Adverse Drug Experience Reports will be prepared and submitted to FDA for review. In addition, a Safety Management Team will be established to review all AEs on a quarterly basis in order to identify any trends or rare serious events that might not be evident from the clinical program along with signal detection for potential new safety concerns. In addition to spontaneous reporting, global literature is regularly screened for AEs.

9 BENEFIT-RISK CONCLUSIONS

C. difficile infection is a potentially life-threatening illness that causes considerable morbidity. Reducing rCDI is the focus of many government-funded initiatives due to its significant impact on public health. For an individual patient, beyond the mortality and morbidity risk it is important to consider how diarrheal illnesses are both debilitating and with significant negative impact on social life. A cycle of recurrences would only multiply such morbidity. Currently, antibiotics are the only first-line therapy approved for treating CDI and recurrences. Antibiotic therapy comes with its own risks associated with an increased likelihood of recurrence due to their negative effects on the gut microbiome. Recurrence of CDI is a significant health problem and there are currently few solutions available to help people who experience rCDI beyond antibiotics.

RBX2660 aims at restoring the healthy gut microbiome and shows clinically meaningful benefit for patients suffering from rCDI. Treatment with RBX2660 resulted in a statistically significant reduction in CDI recurrence compared with placebo in the pivotal, placebo-controlled, Phase 3 study. This result is supported by a totality of evidence of both strong benefits and defined risks of treatment from the open-label studies.

The risks of RBX2660 include diarrhea and abdominal pain, which were the most common AEs observed in the clinical trials. And were predominately mild to moderate.

RBX2660 was well tolerated in clinical trials, with the risks of therapy well-characterized and manageable. The benefit of RBX2660 outweighs these risks while addressing an urgent unmet medical need for treatment of a serious rare disease. RBX2660 provides a new option for patients in need of treatment for rCDI, thereby breaking the cycle of disease recurrence.

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11 APPENDICES

11.1 Adaptive Design Report for the Pivotal Phase 3 Study 2017-01

Berry Consultants
Statistical Innovation

Adaptive Design Report for the Phase 3 Trial (2017-01) for Evaluating the Efficacy and Safety of Rebiotix RBX2660 (microbiota suspension) for the Treatment of Recurrent Clostridium Difficile Infection

Submitted to Rebiotix

August 7, 2019

Introduction

This document describes the adaptive design for a randomized clinical trial to support administration of Rebiotix RBX2660 (microbiota suspension) via enema for the prevention of recurrent *Clostridium difficile* infection (CDI). The trial will enroll and treat up to 270 subjects with recurrent CDI and includes up to two interim analyses at a minimum of 160 and 220 subjects complete (i.e., completed the 8-week follow-up assessment and/or completion of Treatment Outcome form after receiving the blinded treatment). The first interim analysis may occur after 160 subjects complete, pending FDA review of the proposed adaptive design. Early trial success or failure may be declared at either of the interims depending on the strength (either positive or negative) of the observed data. The primary analysis of the trial will be a Bayesian hierarchical model, which formally incorporates data from a previous randomized Phase 2b study (Protocol 2014-01) of RBX2660.

Treatment Arms

Subjects will be randomized to one of the following treatments:

- Treatment: A single dosing of RBX2660 via enema
- Control: A single dosing of placebo via enema

A 2:1 randomization ratio will be used to incentivize patient enrollment in the trial.

Primary Analysis Population

The primary analysis will be conducted using the Modified Intent-to-Treat (mITT) population described in the statistical analysis plan (SAP). Subjects in the mITT population who withdraw due to CDI related symptoms will be counted as treatment failures. Up to 270 patients will be treated in the trial. Patients in the mITT population who dropout prior to completion will be treated as failures; the effect of dropout is described in the section **Operating Characteristics Under Dropout**.

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Primary Endpoint

The primary endpoint of the trial is recurrence of CDI within 8 weeks of blinded treatment. Treatment success is defined as the absence of CDI diarrhea through 8 weeks after completing the blinded study treatment.

Statistical Model

Historical Data

The primary statistical analysis will use a hierarchical model to dynamically borrow information about the treatment effect from the previous Phase 2b trial (Protocol 2014-01). The efficacy objectives of the previous study were to assess the effectiveness of two dosing strategies of RBX2660 in patients with CDI. In one dosing group, patients received two enemas of RBX2660 (administered 7± 2 days apart). In the alternate dosing group, patients received a single enema of RBX2660 along with a placebo (administered 7± 2 days apart). Both treatments were compared to patients in a control arm who received 2 administrations of placebo via enema (administered 7± 2 days apart). Data from this trial are shown in Table 1.

Table 1: *Data from Phase 2b trial (Protocol 2014-01).*

Group	Dosing	# Subjects	# Responders	Response Rate
A	2 doses of RBX2660	41	25	0.610
B	Placebo	44	19	0.432
C	1 dose of RBX2660	42	25	0.595

The hierarchical model will incorporate data from groups B and C, excluding group A due to the difference in treatment dosage.

The patients enrolled in this study are expected to be enrolled from a population similar to the Phase 2b trial. In the case that subjects from the Phase 2b trial differ substantially from the current study, the proposed borrowing strategy allows for heterogeneity between study populations and dynamically borrows less information when the data from the new study differs from the Phase 2b study.

For example, one difference between the studies is that patients with a single recurrence of CDI may be enrolled in this trial whereas they were excluded from the Phase 2b trial. We expect that this change will minimally impact the baseline rate of CDI recurrence observed in the new study. However, if we did observe a control response rate that differed substantially from the Phase 2b study, the design would borrow less information, reducing the role of the historical data in our inference. This effect is described in the section **Strength of the Borrowing**.

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Hierarchical Model

The data from the Phase 3 trial and Phase 2b trial will be analyzed using a Bayesian hierarchical model. The analysis will be conducted using a modified intent to treat population. Let $N_{k,s}$ be the number of subjects assigned to treatment k (k = T for patients who receive a single dose of RBX2660 and k = C for patients who receive the control) in study s ($s = 1$ for the current Phase 3 study, $s = 2$ for the previous Phase 2 study). We model the number of responders, $X_{k,s}$, in each arm/study as

$$X_{k,s} \sim \text{Binomial}(N_{k,s}, p_{k,s})$$

where $p_{k,s}$ is the underlying event rate for arm k in study s. The event rates are transformed to the log-odds scale and modeled as:

$$\log\left(\frac{p_{C,s}}{1-p_{C,s}}\right) = \alpha_s$$

for the control arms and

$$\log\left(\frac{p_{T,s}}{1-p_{T,s}}\right) = \alpha_s + \theta_s$$

for the treatment arms. The parameter θ_s represents the effect of RBX2660, relative to placebo, on the log-odds scale for trial s. Hierarchical models are used to borrow information about the treatment and control effects across studies. The following prior is used for the control rates across the two trials:

$$\alpha_s \sim N(\alpha, \tau_\alpha^2) \quad \text{for } s = 1, 2$$
$$\alpha \sim N(0, 10^2)$$
$$\tau_\alpha^2 \sim \text{Inverse Gamma}(0.001, 0.1)$$

A similar prior is used for the treatment effects across the two trials:

$$\theta_s \sim N(\theta, \tau_\theta^2) \quad \text{for } s = 1, 2$$
$$\theta \sim N(0, 10^2)$$
$$\tau_\theta^2 \sim \text{Inverse Gamma}(0.01, 0.01)$$

The priors on α and θ are chosen to be conservative and emphasize a prior assumption of a control response rate near 0.5 with a treatment effect centered around 0. The priors on the hierarchical variance terms τ_α^2 and τ_η^2 are parameterized by their location μ and weight ω (rather than the traditional $shape = \omega/2$ and $scale = \mu^2 \cdot \omega/2$ parameterization). These priors were chosen to encourage dynamic borrowing between the studies. This distribution is defined by the density:

$$f(x|\mu, \omega) \propto \frac{e^{-\mu^2\omega/2x}}{x^{\omega/2+1}}$$

The selection of the borrowing hyper parameters is discussed in the **Strength of the Borrowing** Section of this design report.

Success Criteria

Our primary interest lies in the treatment effect (TE) for the current Phase 3 study (s = 1):

$$TE = p_{T,1} - p_{C,1}$$

The primary goal of the trial is to demonstrate the efficacy of RBX2660 by testing the hypothesis:

$$H_0: TE \leq 0 \qquad \text{vs.} \qquad H_A: TE > 0$$

We test this hypothesis by calculating the posterior probability of superiority, Pr($TE > 0$ | Data) , which is equivalent to Pr($\theta_1 > 0$ | Data). The thresholds for defining success at each analysis are summarized in Table 2.

Table 2: *Success criteria at each analysis. The * indicates that the success criteria may be updated at the final analysis to ensure the correct amount of α −spend.*

Analysis	Criteria	Subjects complete
Interim 1	Pr($TE > 0$ | Data) > 0.99943	Minimum 160
Interim 2	Pr($TE > 0$ | Data) > 0.99943	220
Final	Pr($TE > 0$ | Data) > 0.99943*	Maximum 270
Final	Pr($TE > 0$ | Data) > 0.97706	Maximum 270

The success criteria of 0.99943 was chosen using a Pocock spending function for analysis performed at 160, 220, and 270 patients complete with a cumulative α spend of 0.00125. Success at this level is considered appropriate for approval based on results from a single trial.

Uncertainty from several factors make assessing the exact α −spend at each analysis difficult to quantify. First, the definition of the mITT population allows for the final analysis to be performed with fewer than 270 complete subjects. In addition, the first interim analysis may be performed when more than 160 subjects are enrolled and treated. Finally, the second interim analysis may be omitted if the first interim analysis occurs sufficiently close to 220 subjects enrolled. Therefore, to account for this uncertainty, the success criteria at the final analysis will be updated to reflect the information spend at each interim analysis and ensure a cumulative α spend of 0.00125. This change will be made based on the fraction of information at each interim analysis and does not depend on the outcome data for the trial.

The lower, secondary, threshold of 0.97706 at the final analysis allows for success at the less demanding $\alpha = 0.025$ level. The second threshold at the final analysis controls the overall Type I error rate for the trial at 2.5% (without borrowing), accounting for the α spend at the interims and higher look at the final analysis.

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Futility Stopping

The trial will stop for futility at either interim if the predictive probability (PP) of success at the final analysis is sufficiently small. The predictive probabilities will be computed using a final threshold of 0.97706 and assuming a final enrollment of 270 subjects. Bayesian predictive distributions incorporate the uncertainty around the model parameters as well as the uncertainty in the outcomes of subjects not yet enrolled. The trial will stop for futility if there is limited probability that the trial will reach the success criterion, even at the maximum sample size. The criteria for declaring futility at each interim are shown in Table 3.

Table 3: *Futility criteria at each analysis.*

Analysis	Criteria	Subjects complete
Interim 1	PP(Success) < 0.01	Minimum 160
Interim 2	PP(Success) < 0.01	220

Strength of the Borrowing

The hyper-priors on τ_α^2 and τ_θ^2 are chosen to encourage dynamic borrowing from the Phase 2b trial. Dynamic borrowing is the idea that when the observed data in the current trial is similar to the previous trial, the strength of our conclusions should increase. On the other hand, if the newly observed data is quite different from the previous data, the prior data should contribute less information to our inference.

Consider two extremes. The first is the case of full borrowing, where data from the previous study is added to data from the new trial and inference is performed using the full weight of both studies. Alternatively, we could do no borrowing, ignoring information from the previous trial and conducting our analysis with only the new information. The hyper-priors on τ_α^2 and τ_θ^2 are specified to provide dynamic borrowing, behaving similarly to the full borrowing approach when the observed data resembles the Phase 2b data and mimicking the no borrowing approach when the results differ in the two trials. The effect of the dynamic borrowing is illustrated in Figure 1.

Figure 1: *Effective sample size (ESS) borrowed on the treatment arm for different observed treatment and response rates at the first interim.*

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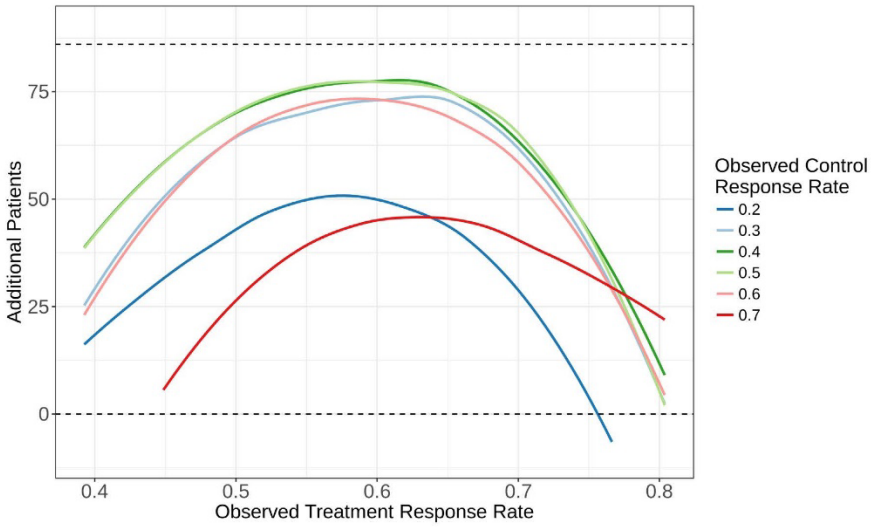

Figure 1 summarizes the amount of borrowing from the hierarchical model for different observed treatment success proportions ($X_{T,1}/N_{T,1}$, the x-axis) and different observed control proportions ($X_{C,1}/N_{C,1}$, differently colored lines). At the first interim analysis, 104 subjects are assumed to be enrolled to the treatment arm with 56 subjects enrolled in the control arm. The y-axis shows the effective number of additional treatment patients borrowed by the model, given by the equation

$$\text{Additional patients} = \frac{\text{Var}(\theta_{full})}{\text{Var}(\theta_1)}(N_1 + N_2) - N_2$$

The quantity $\text{Var}(\theta_1)$ is the posterior variance of the treatment effect under the hierarchical model and $\text{Var}(\theta_{\text{full}})$ is the variance of the treatment effect under the full borrowing model. The ratio of these terms is the relative effective sample size, which measures the uncertainty in the hierarchical model relative to the full model. When this ratio is small, there is substantially more uncertainty in the hierarchical model compared to the full model.

The horizontal dashed lines correspond to 0 (no borrowing from the Phase 2b data) and 86 (full borrowing of the Phase 2b data of Groups B and C). The hierarchical model borrows the most patients from the Phase 2b trial when the observed treatment response rate is close to the treatment response rate in the previous study (a rate of 0.595). As the observed control rate differs from the historical rate, fewer additional patients from the previous trial are used and the amount of borrowing diminishes. When the observed control rate differs from the historical rate, borrowing is strongest when the log-odds of the treatment effect are similar to the phase 2b data. The chosen hyper-priors create appropriate dynamic behavior, decreasing uncertainty when then observed data is in concordance with the previous trial and increasing uncertainty when the Phase 3 trial results differ from the Phase 2b trial.

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Example Trials

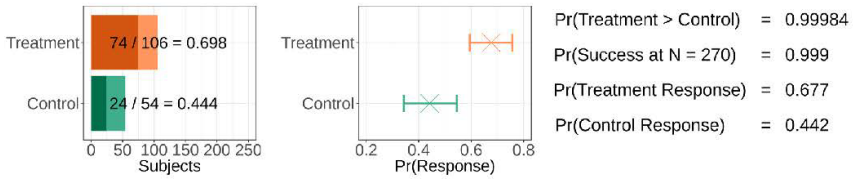
This section contains several example trials selected to highlight aspects of the analysis and design. For each example trial result, we show the following information at each analysis.

- **Observed data**: a bar plot showing the number of patients with complete data enrolled per arm (total width of the bar) as well as the number of responders (darker shaded region).
- **Credible intervals**: plot showing the fitted response rate for each arm (the X) and 95% credible interval for each arm.
- **Pr(Treatment > Control)**: the posterior probability of a positive treatment effect in the Phase 3 trial, used for declaring trial success according to Table 2.
- **Pr(Success at N = 270)**: the predictive probability of success at the final analysis. The trial stops for futility when this probability drops below 0.001.
- **Pr(Treatment Response)**: posterior mean of the treatment arm response probability
- **Pr(Control Response)**: posterior mean of the control arm response probability

Example Trial 1

The first interim occurs when the minimum of N=160 subjects have complete data (i.e., completed their 8-week follow-up assessment and/or completion of Treatment Outcome form after receiving the blinded treatment). Example 1.1 shows the status of the trial at the first interim.

Example 1.1: *Status of the trial at the 1st interim*



The response rate on the control arm is 44.4%, which is similar to the Phase 2b data, and the response rate on the treatment arm is 69.8%, which is larger than observed in the Phase 2 trial. The posterior estimates of the treatment response rate are smaller than the observed rate due to the borrowing from the Phase 2b data. The posterior probability of a positive treatment effect exceeds 0.99943 and success is declared.

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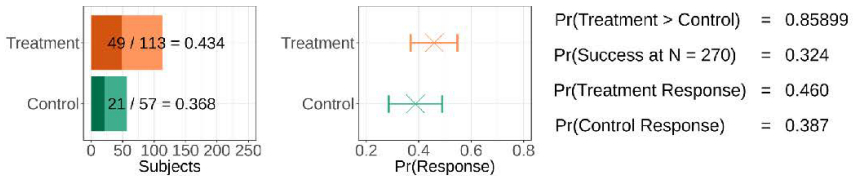
Example Trial 2

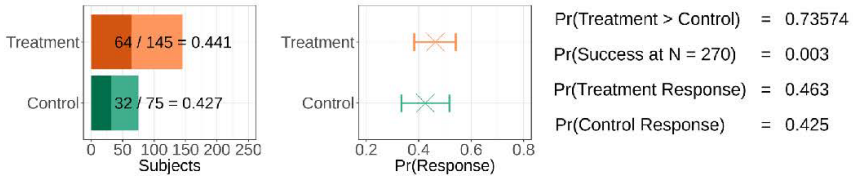
The first interim occurs when N=170 subjects have complete data. Example 2.1 shows the status of the trial at the first interim.

Example 2.1: *Status of the trial at the 1st interim*



The observed response rates in this scenario are much smaller than in the Phase 2b data, with an observed treatment effect of 0.066. While the predictive probability of success is small (0.324), it is sufficient to continue the trial to the second interim.

Example 2.2: *Status of the trial at the 2nd interim*

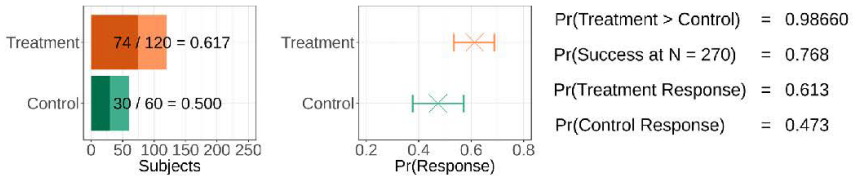


The second interim, shown in Example 2.2, occurs when 220 patients have complete data. The observed response rates have increased in both arms, though the observed treatment effect is smaller (0.014). While the estimated treatment effect is positive, the probability of declaring success at the final analysis is 0.003 which is sufficiently small to stop the trial for futility.

Example Trial 3

In this example, the first interim analysis occurs when 180 subjects have been enrolled. The observed control rate is 0.5, somewhat higher than seen in the Phase 2b trial. Example 3.1 shows the status of the trial at the first interim

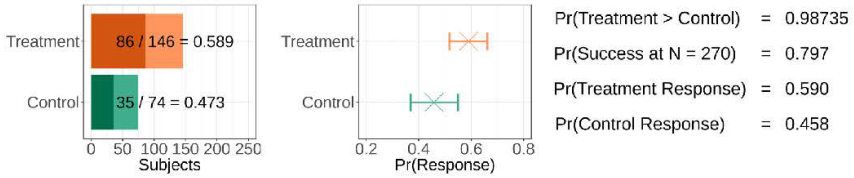
Example 3.1: *Status of the trial at the 1st interim*



The posterior estimate of the control response rate (0.473) is somewhat lower than

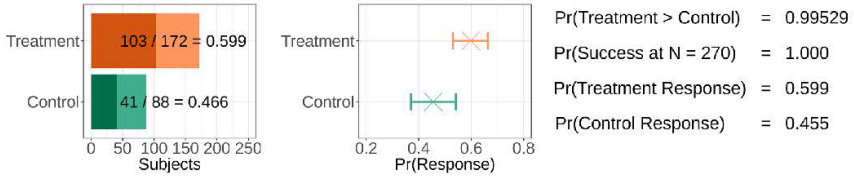
observed due to the borrowing and the posterior probability of a treatment effect is insufficiently high to declare success at the first interim.

Example 3.2: *Status of the trial at the 2nd interim*



At the second interim (N=220, shown in Example 3.2), the observed response rates in both arms have been reduced. The trial has not shown sufficient evidence to declare success or futility and continues enrollment, though the predictive probability of success at the final analysis remains optimistic (0.797).

Example 3.3: *Status of the trial at the final analysis*



The final analysis is conducted when 270 patients have completed follow-up and 260 subjects qualify for the mITT population (Example 3.3). The single trial success criteria is updated to 0.99931, using the information fraction and 0.99943 criteria at the first two interims (0.692 and 0.846) to ensure an overall α −spend of 0.00125. The trial is declared a success at the 0.025 level, though it does not meet the single trial success criteria.

Performance Characteristics

The performance characteristics of the design were determined through trial simulation. We assume several scenarios for the underlying response probability for the control and treatment arms, considering control probabilities in {0.2, 0.3, 0.4, 0.45, 0.5, 0.6, 0.7} and additive treatment effects of {0.0, 0.05, 0.1, 0.15, 0.2, 0.25, 0.3}. The scenario with a control probability of 0.45 and a treatment effect of 0.15 corresponds most closely to what was observed during the Phase 2b trial. The scenarios with treatment effects of 0 correspond to null scenarios. For the purposes of simulation, we assume that patients are randomized in 3 patient blocks, that interim analysis were performed with exactly 160 and 220 subjects completed, and that the 270 mITT subjects have complete data at the final analysis.

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Operating Characteristics

We simulated 1,000,000 virtual trials for each scenario. The results of the simulations are summarized using the following operating characteristics.

- **Pr(Success)**: the cumulative probability of declaring success at the analysis. For example, Pr(Success) under the section Interim 2 includes the probability of declaring success at either Interim 1 or Interim 2. For the final analysis, Pr(Success) (0.99943) and Pr(Success) (0.97706) include the cumulative probability of declaring success at the single trial and lower success levels respectively.
- **Pr(Futility)**: the cumulative probability of declaring futility at the analysis
- **# Subjects**: the average number of subjects enrolled in the trial
- **Bias**: the average error in estimating the true (simulated) treatment effect.

Table 4 shows an example set of operating characteristics when the simulated control response probability is 0.45.

Table 4: *Operating characteristics of the design when the underlying control response probability is 0.45.*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.001	0.445	0.001	0.732	0.002	0.042
0.05	0.006	0.204	0.012	0.424	0.018	0.204
0.1	0.037	0.065	0.072	0.158	0.106	0.528
0.15	0.141	0.014	0.257	0.036	0.354	0.833
0.2	0.363	0.002	0.564	0.005	0.695	0.969
0.25	0.651	0.000	0.838	0.000	0.919	0.997
0.3	0.868	0.000	0.965	0.000	0.989	1.000

Under the null scenario where both the control and treatment arms have a 45% true response rate, 45% of trials stop for futility at the first interim analysis (N = 160) and 0.1% of trials meet the success criterion. By the second interim, 73% of trials have stopped for futility. Assuming a final threshold of 0.97706, the probability of trial success at or before the final analysis is 0.042. The type I error of this trial is inflated beyond the nominal 2.5% level because of the borrowing from the positive phase 2 data.

In the scenario with a treatment effect of 0.15 (which most closely reflects the observed data from Phase 2), the trial has 83% power. Broken down by analysis, 14% of trials declare success at the first interim, another 12% of trials declare success at the second interim, 9% of trials reach the higher success threshold of 0.99875 at the final analysis and 48% of trials declare success at the lower hurdle at the final analysis. When the true

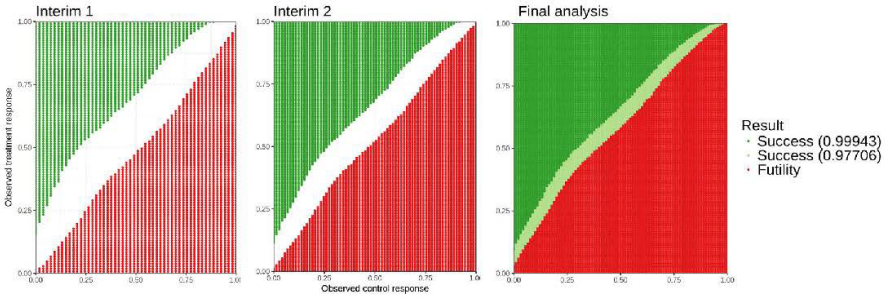
effect is 0.10, the overall power drops to 52% with 77% of trials progressing to the final analysis.

Operating characteristics for simulations with control response probabilities in {0.2, 0.3, 0.4, 0.5, 0.6, 0.7} are included at the end of this section.

Decision Rules

The decision rules of the trial at each analysis can be characterized in terms of the observed effect sizes (# responders / # subjects enrolled) in each arm. The effects leading to either futility or success, colored by result, are shown in in Figure 2.

Figure 2: *Scatterplots of the observed response rates for both arms, colored by result.*



The design has a clear decision boundary at the first interim, declaring futility or success when the observed treatment effect is sufficiently large or small. The gap between success and futility narrows at the 2nd interim, as more information about the arms informs decision making. The effect size required to win is smallest at the final interim.

Operating Characteristics with No Borrowing

For comparison we include the operating characteristics of an alternate design without borrowing. This design uses the same interim schedule as the borrowing design but does not include stopping for futility. Table 6 compares the cumulative probability of declaring success for at each interim for both designs when the simulated control response rate is 0.45.

Table 6: *Operating characteristics for the borrowing design (borrow) and alternate design (none) at each interim analysis when the simulated response rate is 0.45.*

	Interim 1		Interim 2		Final (0.99943)		Final (0.97706)	
Effect	Borrow	None	Borrow	None	Borrow	None	Borrow	None
0	0.001	0.000	0.001	0.001	0.002	0.001	0.042	0.022
0.05	0.006	0.004	0.012	0.007	0.018	0.010	0.204	0.106
0.1	0.037	0.019	0.072	0.038	0.106	0.059	0.528	0.320
0.15	0.141	0.072	0.257	0.141	0.354	0.212	0.833	0.629
0.2	0.363	0.202	0.564	0.362	0.695	0.496	0.969	0.877
0.25	0.651	0.429	0.838	0.655	0.919	0.794	0.997	0.977
0.3	0.868	0.693	0.965	0.883	0.989	0.955	1.000	0.998

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For a simulated treatment effect of 0.15, the borrowing design has 20% more power than the alternate design. Broken down by interim, the borrowing design is 7% more likely to declare success at the first interim, 12% more likely to declare success by the second interim, and 14% more likely to reach the higher threshold by the final analysis. The borrowing design is more likely to declare success when the simulated treatment effect is 0, with a 4.2% chance of declaring success compared to 2.2% for the alternate design.

Operating Characteristics Under Drop Out

Per the protocol, subjects who drop out of the mITT population prior to the final analysis for CDI related symptoms will be treated as treatment failures (regardless of arm). Therefore, simulated dropout has the practical effect of reducing the simulated control and treatment rates and the tables in the following second can be used to understand the performance of the design under a range of dropout scenarios.

For example, suppose that the true response rate on the control arm was 0.45, the true treatment effect was 0.20 and the dropout rate was 10%. Then, this trial would have the same operating characteristics of a trial with a simulated 0.405 control rate, a 0.145 treatment effect, and no dropout. The operating characteristics in this scenario can be adequately approximated using Table 8.

Additional Operating Characteristics

This section contains operating characteristics for control response probabilities in {0.2, 0.3, 0.4, 0.5, 0.6, 0.7}. A description of these tables can be found under the operating characteristics section.

Table 6: *Operating when the underlying control response probability is 0.2*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.000	0.462	0.001	0.732	0.001	0.050
0.05	0.003	0.225	0.009	0.449	0.015	0.203
0.1	0.015	0.087	0.044	0.208	0.077	0.466
0.15	0.054	0.026	0.146	0.070	0.241	0.740
0.2	0.153	0.005	0.353	0.015	0.515	0.919
0.25	0.346	0.001	0.629	0.002	0.790	0.986
0.3	0.608	0.000	0.862	0.000	0.949	0.999

Table 7: *Operating when the underlying control response probability is 0.3*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.000	0.539	0.000	0.811	0.001	0.025
0.05	0.002	0.301	0.004	0.571	0.007	0.114
0.1	0.010	0.122	0.026	0.285	0.044	0.347
0.15	0.049	0.033	0.119	0.089	0.189	0.679
0.2	0.172	0.006	0.353	0.016	0.495	0.912
0.25	0.419	0.001	0.674	0.002	0.812	0.988
0.3	0.710	0.000	0.901	0.000	0.964	0.999

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Table 8: *Operating when the underlying control response probability is 0.4*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.000	0.506	0.001	0.788	0.001	0.027
0.05	0.003	0.246	0.007	0.495	0.011	0.152
0.1	0.023	0.084	0.050	0.204	0.076	0.454
0.15	0.103	0.019	0.199	0.051	0.286	0.784
0.2	0.294	0.003	0.489	0.007	0.628	0.955
0.25	0.575	0.000	0.790	0.001	0.891	0.995
0.3	0.827	0.000	0.951	0.000	0.984	1.000

Table 9: *Operating when the underlying control response probability is 0.5*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.001	0.389	0.002	0.668	0.003	0.063
0.05	0.010	0.166	0.019	0.358	0.027	0.260
0.1	0.054	0.049	0.101	0.123	0.143	0.595
0.15	0.187	0.009	0.316	0.025	0.418	0.869
0.2	0.431	0.001	0.626	0.003	0.748	0.978
0.25	0.700	0.000	0.870	0.000	0.938	0.998
0.3	0.885	0.000	0.971	0.000	0.992	1.000

Table 10: *Operating when the underlying control response probability is 0.6*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.004	0.296	0.006	0.559	0.007	0.108
0.05	0.022	0.117	0.038	0.269	0.052	0.351
0.1	0.089	0.035	0.153	0.089	0.205	0.671
0.15	0.241	0.007	0.384	0.019	0.490	0.894
0.2	0.475	0.001	0.670	0.003	0.789	0.981
0.25	0.733	0.000	0.894	0.000	0.957	0.999
0.3	0.925	0.000	0.986	0.000	0.998	1.000

Table 11: *Operating when the underlying control response probability is 0.7*

	Interim 1		Interim 2		Final analysis	
Effect	Pr(Success)	Pr(Futility)	Pr(Success)	Pr(Futility)	Pr(Success) (0.99943)	Pr(Success) (0.97706)
0	0.004	0.290	0.007	0.555	0.009	0.108
0.05	0.021	0.120	0.036	0.279	0.049	0.325
0.1	0.082	0.033	0.141	0.086	0.197	0.661
0.15	0.246	0.005	0.396	0.013	0.529	0.919
0.2	0.559	0.000	0.760	0.001	0.880	0.995
0.25	0.877	0.000	0.977	0.000	0.995	1.000
0.3	1.000	0.000	1.000	0.000	1.000	1.000

Computational Details

Simulations were run using R (R Core Team 2017) version 3.4.3. Bayesian computations were performed using the Stan (Stan Development Team 2018) version 2.17.2.

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Many samples from the posterior distribution are required to determine whether the posterior probability is greater than 0.99943 with sufficient precision. For the power calculations, for each trial we ran a single MCMC sampler for 10,000 iterations. When executing the trial, if the posterior probability of a positive treatment effect is close to the boundary we recommend using more samples to obtain sufficient precision. For example, using 1,000 independent Markov chains with 10,000 post burn-in iterations each.

11.2 Pivotal Phase 3 Study 2017-01: Summary of Results for Other Efficacy Endpoints

Results for other efficacy endpoints (in addition to the primary and secondary endpoints) are summarized in Table 29.

Table 29: Pivotal Phase 3 Study 2017-01 – Results for Other Efficacy Endpoints (mITT)

Baseline Characteristics Predictive of Efficacy Outcomes of Single Dose RBX2660
• Although all interactions of the covariates with treatment group showed no statistically significant impact (at the alpha level of 0.05) on Treatment Success after blinded treatment, a multivariate logistic regression analysis was also performed to identify predictors (if any) for Treatment Success with one RBX2660 dose vs. those who were unsuccessful. • In this analysis which combined both blinded and open-label outcomes for one dose of RBX2660, age group had a statistically significant covariate effect (p=0.0396). • No other covariates or interactions showed a statistically significant impact on Treatment Success rate.
Fecal Microbiome Composition During the Blinded Period
• Fecal sequencing analysis showed that the microbiome composition of patients adjudicated as Treatment Success was significantly shifted by 1 week after study treatment to more closely resemble the RBX2660 composition, and this shift continued through at least 6 months post-treatment. • The composition of all post-treatment time points was significantly different than Baseline ($p < 0.001$, Generalized Wald Test) and was primarily characterized by increased Bacteroidia- and Clostridia-class bacteria concomitant with decreased Gammaproteobacteria- and Bacilli-class bacteria after treatment. • The majority of Treatment Failures were censored from this analysis due to failure prior to the earliest prescribed time point at 1 week.
Charlson Comorbidity Index Throughout the Study
• Treatments with both RBX2660 and Placebo are associated with reduced comorbidity throughout the study with a higher trend for reduced comorbidity in the RBX2660 arm. • The mean (SD) CCI scores in the mITT population at Screening and Week 8 post-blinded treatment were 2.9 (2.33) and 2.2 (1.95) for Placebo (n=54), and 3.7 (2.95) and 2.8 (2.31) for RBX2660 (n=117), respectively.
Health-Related Quality of Life
• The mean (SD) health-related quality of life Cdiff32 scores in the mITT population at the Screening and Week 8 post-blinded treatment were 44.61 (19.88) and 60.9 (24.82), respectively, for Placebo (n=83), and 45.84 (17.74) and 67.84 (22.02), respectively, for RBX2660 (n=174).
ATLAS score for CDI severity of qualifying CDI event
• The ATLAS scores were comparable between RBX2660 and Placebo arms with a mean (SD) total ATLAS score of 2.9 (0.99) for the SP population
Recurrence of CDI within 8 Weeks of Open-Label Treatment
• In the mITT population, for patients treated with Placebo and patients who documented Placebo failure and then went on to receive an open-label RBX2660 enema, the Treatment Success rates were 62.4% and 62.5%, respectively.

Recurrence of CDI within 8 Weeks of Blinded or Open-Label RBX2660 Treatment
• Treatment Success rates were 71.2% in patients treated with blinded RBX2660 and 53.7% in patients who documented blinded RBX2660 failure and then went on to receive an open-label RBX2660 enema, in the mITT population. • By combining the blinded and open-label efficacy results, an increased Treatment Success rate of 83.6% (148/177) is observed; 22 patients received a total of 2 doses of RBX2660 through the combined dosing in the blinded and open-label portions of the study.
Occurrence of CDI through 6 Months after One RBX2660 Enema
• Treatment with one RBX2660 enema (blinded or open-label) demonstrated a Sustained Clinical Response rate of 131 (92.9%), 131 (92.9%), and 125 (92.6%) through 6 months of treatment in mITT, ITT, and PP populations, respectively.
Concentration of Vancomycin-Resistant Enterococcus (VRE) in Stool Samples for Patients Who Were Carriers at Baseline
• A total of 183 patients (68.5%, 183/267) participated in the patient sampling program, and a total of 903 stool samples were received. Samples received during the blinded treatment were included in this analysis, totaling 717 samples. • Thirty samples (4.2%, 30/717) tested positive for VRE by the culturing assay. Eight (5%, 8/161) of the Baseline samples tested positive for VRE, slightly rose to 10 (6.5%, 10/155) samples by Week 1, then numbers gradually decreased over time to 1 (1%, 1/83) VRE-positive sample at Month 6. • Of note, 5 patients had a positive VRE test at Baseline and were negative after treatment, of which 4 were in the RBX2660 treatment group.
Presence of C. difficile in Stool Samples Throughout the Study

• Presence of *C. difficile* in stool was measured in all samples received (ie, patients randomized to either RBX2660 or Placebo, as well as patients in the open-label period) by 3 tests (*C. difficile* Toxin B PCR [CDBPCR], Quik Chek Antigen, and Quik Chek Toxin), of which CDBPCR test was of highest sensitivity and Quik Chek tests were of highest specificity.
• The pattern of results for the presence of *C. difficile* in stool samples that was observed for the 3 different tests were similar, with an increase in the percentage of positive tests at Week 1 and Week 4 after treatment, with a decrease thereafter.
• The number of *C. difficile*-positive samples then rose to 67 (43%, 67/154) of the Week 1 samples, 53 (45%, 53/117) Week 4 samples, and then numbers gradually decreased over time to 13 (16%, 13/82) of the Month 6 samples.
• The percentages of positive samples differed among tests, with the CDBPCR test showing the highest percentage of positive samples, as would be expected since it has the highest sensitivity.
  ○ Of the 903 received samples, 318 (35%) tested positive for *C. difficile* by the CDBPCR.
  ○ Of the 717 samples from the blinded phase, 225 (31%) tested positive.
  ○ Twenty-nine (18%, 29/161) Baseline samples tested positive for *C. difficile* with this assay.

11.3 Summary of Safety in Phase 2 Study 2013-001

A summary of AEs occurring in the open-label, first in human Phase 2 Study 2013-001 is provided in Table 30.

Table 30: Phase 2 Study 2013-001 – Overview of Adverse Events

Safety through 6-months after last treatment	**RBX2660 N = 34 n (%)**
All AEs	28 (82.4%)
Number of AEs	188
AEs by maximum severity	
Mild	25 (73.5%)
Moderate	13 (38.2%)
Severe	5 (14.7%)
Potentially life-threatening	1 (2.9%)
All SAEs	7 (20.6%)
Deaths*	1 (2.9%)

*Additional information on patient deaths, including the one reported in Study 2013-001, is provided in Section 7.5.6 and Appendix 11.5.

11.4 Integrated Safety Population: Baseline Patient Demographics and Characteristics

Baseline demographics and characteristics for patients in the integrated safety population are summarized in Table 31.

Table 31: Integrated Safety Population – Baseline Demographics and Characteristics (ITT Analysis Set)

	Placebo Only (N = 83)	RBX2660 Only (N = 763)	Placebo / Open-Label RBX2660 (N = 48)	RBX2660 / Open-Label RBX2660 (N = 167)	All RBX2660 (N = 978)
Age (years), mean (SD) Min, max	58.1 (16.48) 19.0, 90.0	61.4 (17.60) 18.0, 103.0	58.0 (18.19) 24.0, 92.0	62.8 (18.34) 18.0, 93.0	61.5 (17.77) 18.0, 103.0
Female, n (%)	60 (72.3%)	516 (67.6%)	30 (62.5%)	111 (66.5%)	657 (67.2%)
White, n (%)	75 (90.4%)	713 (93.4%)	46 (95.8%)	158 (94.6%)	917 (93.8%)
Duration of CDI (days), mean (SD)	24.3 (13.98)	32.3 (27.35)	25.7 (16.23)	32.3 (27.04)	31.9 (26.85)
Number of Previous Episodes of CDI ≥ 1 1-3 ≥ 3	 83 (100.0) 60 (72.3) 57 (68.7)	 757 (99.2) 450 (59.0) 595 (78.0)	 48 (100.0) 23 (47.9) 40 (83.3)	 167 (100.0) 98 (58.7) 133 (79.6)	 972 (99.4) 571 (58.4) 768 (78.5)
Hospitalization					
Due to CDI episode, n (%)	17 (20.5%)	106 (13.9%)	5 (10.4%)	25 (15.0%)	136 (13.9%)
Duration (days), median (IQR)	5.0 (3.0)	5.0 (4.0)	5.0 (6.0)	7.5 (7.5)	5.0 (4.0)

11.5 Case Narratives for Patient Deaths

Case narrative (vignettes) for the 18 patients in the integrated safety population experiencing AEs within 6 months of last RBX2660 treatment leading to death are provided in Table 32.

Note that, within the narratives presented in this section, calculation of study days for events is based on time since first exposure to study treatment, while in the discussion of patient deaths in the main text of this document, study days are calculated from time since last treatment.

Table 32: Case Narratives for Patient Deaths in All RBX2660-Treated Patients

Patient	Brief Vignette
2017-01 79 y.o. White Female *Blinded:* RBX2660 Day 1 *Open-label:* RBX2660 Day 29	**Relevant past medical history:** Diabetes mellitus Type 2, Cardiac failure congestive, Cerebrovascular disease, Chronic kidney disease. **Relevant Interim AEs:** Urinary tract infection (moderate, Day 72-76 and Day 122-128, Hypotension (SAE, Hospitalization, severe, Day 126-135 attributed to *C. difficile*), Cardiac failure congestive (SAE, Hospitalization, severe, Day 150-154), Chronic obstructive pulmonary disease (moderate, Day 150-ongoing), Atrial fibrillation (moderate, Day 151-154), and Decubitus ulcer (severe, Day 152-ongoing). **AE resulting in Death: Multimorbidity** (SAE, Disability/Life-threatening, Day 179). The patient had CDI (SAE, Hospitalization, moderate, Day 160-168) with AEs of Urinary tract infection (severe, Day 160-169), Decubitus ulcer (moderate, Day 163-ongoing). Treatment included IV fluids, ceftriaxone, oral vancomycin, fidaxomicin, IV metronidazole, and oral doxycycline. On Day 179 she died. Autopsy report and death certificate not available.
2017-01 75 y.o. White Male *Blinded*: 1 RBX2660 Day 1	**Relevant past medical history:** Cerebrovascular accident, Atrial fibrillation, Parkinson's disease, Coronary artery bypass, Dementia, Epilepsy. **Relevant Interim AEs:** None. **AE resulting in Death: Cardio-respiratory arrest** (SAE, Life-threatening, Day 37). The patient had increasing lethargy over several weeks with Fever (moderate, possible related, Day 27-28) treated with paracetamol, and abdominal pain (moderate, possible related, Day 36-37). On Day 37 he was found unresponsive by caregiver and transported to emergency room; cardiopulmonary resuscitation was not successful. No autopsy was performed.
2014-01 88 y.o. White Male *Blinded:* RBX2660 Day 1 and Placebo Day 9	**Relevant past medical history:** Anemia, Peripheral vascular disease, Diabetes Type 2, Below the knee amputation, Chronic obstructive pulmonary disease, Diastolic dysfunction, . **Relevant Interim AEs:** Fall (mild, Day 26-27). **AE resulting in Death: General physical health deterioration** (SAE, Death/Life-threatening, Day 56 The patient had a below-knee amputation Day —6. He had worsened Anemia (SAE, Hospitalization/Disability/Medically important, severe, Day 43-49) treated with transfusion Day 47. Abdominal CT scan revealed right lateral wall Subcutaneous hematoma (mild, Day 47-ongoing; GI work-up was not completed because hematocrit stabilized. He had General health deterioration prompting transition to palliative care and he died on Day 58.

Patient	Brief Vignette
2014-01 83 y.o. White Male *Blinded:* RBX2660 Day 1 and Placebo Day 7	**Relevant past medical history:** Pressure ulcer of feet, Early cellulitis, Coronary atherosclerosis, Congestive heart failure, Chronic atrial fibrillation, Cardiac defibrillator in situ, Cardiac pacemaker, Chronic obstructive pulmonary disease, Prostate cancer, Aspiration pneumonia. **Relevant Interim AEs:** Ischaemic cardiomyopathy (moderate, Day 25-ongoing, Lower gastrointestinal haemorrhage (mild, Day 29-ongoing). **AE resulting in Death: General physical health deterioration** (SAE, Death/Life-threatening, Day 88). On Day 35 the patient underwent aggressive wound care for bilateral heel ulcers and 5th metatarsal head ulcer and chronic osteomyelitis. Bone cultures were positive for methicillin-resistant *S. aureus*, pseudomonas and proteus. and increased white blood cells. On follow-up Osteomyelitis chronic (SAE, Disability, moderate, Day 49-ongoing) was diagnosed and IV vancomycin and cefipime was administered. Chronic obstructive pulmonary disease Day 60-ongoing) was reported. On Day 78 he had ray resection of toe and treatment for bilateral heel osteomyelitis. He was hospitalized for dysphagia, myoclonic jerking movements, and waxing and waning delirium; Parkinsonism (SAE, Hospitalization/Disability/Life-threatening, moderate, Day 88-ongoing) was diagnosed. Aspiration on swallowing evaluation noted. He transitioned to hospice care, had internal defibrillator disconnected, and died on Day 100.
2014-01 87 y.o. White Female *Blinded:* RBX2660 Day 1 and Placebo Day 8 *Open-label:* RBX2660 Day 59 and Day 67	**Relevant past medical history:** Chronic obstructive pulmonary disease, Pulmonary hypertension, Congestive heart failure, Atrial fibrillation **Relevant Interim AEs:** None **AE resulting in Death: Respiratory failure** (SAE, Death/Hospitalization/Disability/Medically important/Life-threatening, Day 223-death) After last open-label RBX2660 on Day 67, the patient had additional recurrences in Aug, Sep and Oct 2015 treated with vancomycin. Abdominal CT on Day 191 revealed colonic mass. On Day 216 underwent transverse colectomy; Adenocarcinoma colon (SAE, Hospitalization/Disability/Medically important/Life-threatening, Day 217-218) was confirmed. Post-op she had Pulmonary oedema (moderate, Day 218-ongoing. On Day 222 she became tachypneic and hypoxic (Respiratory distress, moderate, Day 222-ongoing. On Day 223, 156 days post last RBX2660 dose, she transitioned to comfort care and died.
2014-01 76 y.o. White Male *Blinded:* RBX2660 Day 1 and Day 8	**Relevant past medical history:** Metastatic hormone resistant prostate cancer s/p prostatectomy, Hydronephrosis s/p ureteral stent, Urinary tract infections, Squamous cell lung cancer s/p with left thoracotomy and upper lobectomy, Myocardial infarction, Chronic kidney disease, Follicular lymphoma Grade 1, **Relevant Interim AEs:** Ureteral stent replacement for hydronephrosis complicated by Acute respiratory failure (SAE, Hospitalization/Disability/Medically important/Life-threatening, Day 31-56); Ureteric stenosis (SAE, Hospitalization/Medically important, moderate, Day 31); Arrhythmia supraventricular (moderate), Dyspnoea (moderate), Pneumonia (severe, Day 31-ongoing), Atelectasis (mild, Day 31-43); Hypoxia and Lung infiltration (moderate) and Pulmonary oedema (mild) (both - Day 31-36), Cardiac failure congestive (moderate), Cardiomyopathy acute (moderate), (all - Day 32-ongoing); Chronic obstructive pulmonary disease (moderate, Day 34-ongoing), I. **AE resulting in Death: Acute respiratory failure** (SAE, Death/Hospitalization/Disability/Medically important/Life-threatening, Day 56-death). Acute respiratory failure was reported on Day 56. On Day 74 the site was informed the patient was not eating and had Pyrexia (mild). He was do-not-resuscitate on palliative care and died on Day 75. Death certificate cause: acute respiratory failure with pneumonia; carcinoma in situ of the bronchus and lung as contributing.

Patient	Brief Vignette
2014-01 63 y.o. White Male *Blinded:* RBX2660 Day 1 and Placebo Day 8 *Open-label* RBX2660 Day 31 and Day 36	**Relevant past medical history:** End-stage renal disease, Diabetes, Foot ulcer, Coronary artery disease s/p stent placement, Heart failure, Lacunar infarct. **Relevant Interim AEs:** See below. **AE resulting in Death: Bacteremia, Sepsis, Staphylococcal infection** (SAE, Death/Hospitalization/Disability/Medically important/Life-threatening, Day 60-death), **Respiratory failure** (SAE, Hospitalization/Disability/Medically important/Life-threatening, Day 60-death). On Day 60 the patient became hypotensive and short of breath during dialysis. Intubated; blood Cx positive for methicillin-resistant *S. aureus*; suspected source was dialysis permacatheter or decubitus ulcers. He received multiple antibiotics and surgery was consulted for permacatheter removal. He did not respond to vasopressor support, remained acidotic, was placed on palliative care and died on Day 64. Sepsis attributed to MRSA bacteremia and possibly related to *C. difficile* colitis or healthcare acquired/associated pneumonia.
2014-01 84 y.o. White Female *Blinded:* RBX2660 Day 1 and RBX2660 Day 8 *Open-label:* RBX2660 Day 35 and Day 41	**Relevant past medical history:** Renal insufficiency, Hypertension. **Relevant Interim AEs:** None **AE resulting in Death: Renal failure** (SAE, Death/Hospitalization/Disability/Medically important/Life-threatening, Day 49-death). The patient was hospitalized at treatment initiation. She had increased creatinine (Renal impairment, SAE, Hospitalization, moderate, Day 19-ongoing) and hypokalemia treated with IV fluids and holding antihypertensives. Recurrent *C. difficile* diarrhea on Day 25 was treated with oral vancomycin. Ceftriaxone was administered for *Escherichia* urinary tract infection (mild, Day 26-32). Open-label RBX2660 was administered. On Day 49 hemodialysis was started for Renal failure, Anuria (SAE, Hospitalization/Disability/Medically important/Life-threatening, severe, Day 49-ongoing), and Blood creatinine increased (SAE, Hospitalization/Disability/Medically important/Life-threatening, severe, Day 49-ongoing). She was treated for Urinary tract infection (mild, Day 49-63); diarrhea returned on Day 50 treated with IV metronidazole and oral vancomycin. Diagnosed with Sepsis (SAE, Hospitalization/Disability/Medically important/Life-threatening, moderate, Day 64) due to fever, diarrhea and elevated white blood count. On Day 69 dialysis was discontinued and she died on Day 74.
2014-01 73 y.o. White Female *Blinded:* RBX2660 Day 1 and Day 7	**Relevant past medical history:** Peripheral vascular disease, Above knee amputation, Hypertension, Hypercholesterolemia, Type 2 diabetes. **Relevant Interim AEs:** Constipation (mild, Day 1-ongoing). **AE resulting in Death: Intestinal ischaemia** (SAE, Death, life-threatening, Day 1-Death). The patient missed 6-month call. At late 12-month call on Day 486 she reported ongoing constipation. On Day 738 the site became aware the patient died on Day 564 due to mesenteric ischemia (Intestinal ischaemia). **Included as death due to AEs within 6 months of last RBX2660 in error due to imputation as event onset not in database.**

Patient	Brief Vignette
2013-001 83 y.o. White Female RBX2660 Day 1 and RBX2660 Day 12	**Relevant past medical history:** Chronic obstructive pulmonary disease (O_2 dependent), Macrodantin pulmonary toxicity. **Relevant Interim AEs:** None **AE resulting in Death: Pelvic fracture** (SAE, Death/Hospitalization/Disability/Medically important, Life-threatening, Day 30-47), **Respiratory failure** (SAE, Death/Hospitalization/Disability/Medically important/Life-threatening, Day 44-47). On Day 30 the patient had a Pelvic fracture after a fall following disorientation from a urinary tract infection. She was transferred to a rehabilitation facility. On Day 31 she was hospitalized for respiratory distress, chronic respiratory failure, pelvic fracture, and Urinary tract infection (SAE, Hospitalization/Disability, severe, Day 32-47); she received ceftriaxone, methylprednisolone, pain medication, and physical therapy. On Day 42 she was transferred to rehabilitation facility. On Day 44 she was readmitted for Respiratory failure (hypoxia, chest congestion, CT c/w chronic obstructive pulmonary disease exacerbation due to tracheobronchitis). She was treated with levofloxacin, methylprednisolone, montelukast, nebulizer treatments and oxygen. She continued to decline and was made do-not-resuscitate on Day 45. On Day 47 she desaturated after IV diltiazem, was placed on bilevel positive airway pressure (BiPAP) and morphine drip, and died. The pelvic fracture was reported as the cause of death as it precipitated the exacerbation of respiratory symptoms.
2015-01 94 y.o, White Female RBX2660 Day 1 and Day 8	**Relevant past medical history:** First CDI Day -85 with recurrences Day -73, Day -71, Day -36, Day -23 and Day -9. **Relevant Interim AEs:** None. **AE resulting in Death: *Clostridium difficile* infection (**SAE, Death/Hospitalization/Disability/Life-threatening, severe, Day 21-ongoing) On Day 21 the patient was hospitalized with rCDI (stool positive Day 24). Treatment included metronidazole, vancomycin, FMT via colonoscopy (unknown source), IV fluids and parenteral nutrition. *C. difficile*-positive diarrhea continued and course was complicated by Ileus and Leukocytosis (both - SAE, Hospitalization/Disability/Life-threatening, severe, Day 21-ongoing), Pyrexia (SAE, Hospitalization/Disability/Life-threatening, Day 21-25), Atrial fibrillation (SAE, Hospitalization/Disability/Life-threatening/Medically important, severe, Day 25-ongoing), Acute myocardial infarction (SAE, Hospitalization/Disability, moderate, Day 26-ongoing), and Malnutrition (SAE, Hospitalization/Disability/Life-threatening, moderate, Day 27-ongoing), with increased creatinine and decreased urine output. She transitioned to comfort care and died on Day 31.
2015-01 67 y.o. White Male RBX2660 Day 1 and RBX2660 Day 8	**Relevant past medical history:** Chronic obstructive pulmonary disease, Emphysema, Acute respiratory failure, Chronic respiratory failure, Dyspnoea, Bronchitis, Bronchitis chronic, Lung infiltration, *Mycobacterium kansasii i*nfection. **Relevant Interim AEs:** None. **AE resulting in Death: Cardiac failure congestive** and **Chronic obstructive pulmonary disease** (both - SAE, Hospitalization, severe, Day 118-death). On Day 118, the patient was hospitalized for Cardiac failure congestive, Chronic obstructive pulmonary disease, and Emphysema (SAE, Hospitalization, severe, Day 118-ongoing). Treatment included furosemide, methylprednisolone, ipratropium/salbutamol, albuterol, fluticasone-salmeterol, and tiotropium bromide. He was discharged. On Day 260 he died due to Chronic obstructive pulmonary disease and Cardiac failure congestive.

Patient	Brief Vignette
2015-01 68 y.o. Black Female RBX2660 Day 1 and RBX2660 Day 8	**Relevant past medical history:** Pneumonia, Pulmonary vascular disorder, Acute myocardial infarction, Cardiac failure, Coronary artery disease, Ischaemic cardiomyopathy, Acute respiratory failure, Bronchitis, Chronic obstructive pulmonary disease, Renal failure acute, Renal failure chronic, Urinary tract infection, Cerebrovascular accident, Dementia, Cachexia. **Relevant Interval AEs**: See below. **AE resulting in Death: Sepsis** (SAE, Hospitalization/Disability/Life-threatening, severe, Day 154-death) The patient had multiple hospitalizations including for Acute respiratory distress syndrome (SAE, Hospitalization/Medically important, severe, Day 27-39), Dyspnoea (SAE, Hospitalization/Disability/Life-threatening/Medically important, severe, Day 40-45), Dyspnoea (SAE, Hospitalization/Life-threatening severe, Day 60-66), Dyspnoea (SAE, Hospitalization/Disability/Life-threatening severe, Day 84-ongoing), Chronic obstructive pulmonary disease (SAE, Hospitalization, severe, Day 101-ongoing), She also had Cachexia (SAE, Hospitalization, severe, Day 44-ongoing), Renal failure (severe, Day 78-135), worsening congestive heart failure, non-sustained ventricular tachycardia, and implantable cardioverter defibrillator placement. She was transferred (Day 135) to a skilled nursing facility due to ventilator dependence. On Day 154 she was re-hospitalized for Respiratory distress (SAE, Hospitalization/Disability/Life-threatening, severe, Day 154-ongoing) and Sepsis. Chest x-ray had bilateral pleural effusions and possible right lower lobe infiltrates. Treatment included meropenem, transfusions and nebulizer treatments. ESBL *Klebsiella* was isolated in urine, decubitus wounds, and sputum; CR *Pseudomonas* in urine, sputum and PEG tube; and *Acinetobacter* in wound. On Day 181 respiratory failure continued to worsen; she was DNR and died on Day 185.
2015-01 91 y.o. Black Female RBX2660 Day 1 and RBX2660 Day 8	**Relevant past medical history:** Renal failure chronic, Acidosis, Hypertension, Hyperkalemia, Proteinuria. **Relevant Interim AEs:** See below **AE resulting in Death: Nephropathy** (SAE, Disability/Life-threatening, severe, Day 91-ongoing) On Day 1 the patient was hospitalized for worsening Chronic kidney disease (SAE, Disability, severe, Day 1-ongoing), lower extremity edema and worsening anemia (onset Day -10). Baseline creatinine was 239 µmol/L (44-88). Other subsequent AEs Oedema peripheral (moderate, Day 71-12) and Hypertension (moderate, Day 86-111). Treatment included erythropoietin and antihypertensives. On Day 91 she was hospitalized for Nephropathy. She had worsening hypertension treated with antihypertensives; she refused hemodialysis. Other subsequent AEs included Acidosis (mild, Day 147-ongoing), Hyperkalemia (mild, Day 178-ongoing). On Day 514 she was hospitalized for Bradycardia (SAE, Hospitalization, severe, Day 514-517), Hypertension (SAE, Hospitalization, Day 514-531), Traumatic haematoma (SAE, Hospitalization, severe, Day 514-ongoing) after sustaining facial fractures and retrobulbar hematoma after fall. She underwent surgical repair and was discharged on Day 517. On Day 567 she developed Headache (moderate) and hyperkalemia and was admitted to hospice. She died on Day 619 due Nephropathy.

Patient	Brief Vignette
2015-01 77 y.o. White Male RBX2660 Day 1 and Day 7	**Relevant past medical history:** Lung neoplasm malignant, Chronic obstructive pulmonary disease, Emphysema, *Clostridium difficile* infection, Prostate cancer, Radiotherapy, Arrhythmia, Renal failure chronic, Carotid artery stenosis, . **Relevant Interim AEs:** None. **AE resulting in Death: Death** (SAE, Death, Day 181) On Day 35 the patient was seen in emergency room for Constipation, Duodenal ulcer, Intestinal ulcer, Oesophagitis, White blood cell count increased [all - SAE, Hospitalization, moderate, Day 35-54) and he was hospitalized on Day 36). Abdominal CT showed stomach distention, thickening of the antrum of stomach and duodenum, thickening of rectal wall compatible for proctitis, and vascular changes. Stool was *C. difficile*-positive. Treatment included levofloxacin, vancomycin, and fluconazole. Esophagogastroduodenoscopy and flexible sigmoidoscopy with esophagitis, duodenal and sigmoid ulcers, colitis, proctitis (possibly from prior radiation treatment). He received transfusion, epinephrine injection, and cauterization for persistent duodenal ulcer bleeding. Other AEs included Urinary tract infection (SAE, Hospitalization, Day 45-54), Colitis and Proctitis (both - SAE, Hospitalization, moderate, Day 72-85). On Day 85 gastrointestinal conditions resolved. He died on Day 181; cause was reported as lung cancer, chronic obstructive pulmonary disease, emphysema and colitis.
2019-01 62 y.o. Black Male RBX2660 Day 1 *Retreated* RBX2660 Day 35	**Relevant past medical history:** Cardiomyopathy, Cardiac failure congestive, Atrial fibrillation, Diabetes mellitus, Chronic kidney disease, Obesity, Sleep apnoea syndrome. **Relevant Interim AEs:** Hypoglycemia (severe, Day 71 [30Oct2020]), Cardiac failure (SAE, Hospitalization, severe, Day 91-97) **AE resulting in Death: Cardiac arrest** (SAE, Hospitalization, life-threatening, Day 182) Beginning Day 116 the patient had multiple falls, impaired mobility, gait abnormality, and right-hand weakness. On Day 156 he was admitted with Quadriparesis (SAE, Disability/Hospitalization, severe, Day 115-ongoing). Severe canal stenosis was observed on MRI (C4-C5) and X-ray (C3-C4 with cord compression C5-C6). He underwent cervical discectomy and fusion. He was discharged on Day 162 to a skilled nursing facility. On Day 182 he complained of "stabbing" chest pain with 98/53 mmHg and noted to have heart rate 38 beats per minute, altered mental status, and lethargy. Cardiac arrest was diagnosed by emergency medical services and CPR initiated. He died the same day; an autopsy was not performed.
2019-01 94 y.o. Asian Male RBX2660 Day 1	**Relevant past medical history:** Pneumonia, Pulmonary edema, Cardiac failure congestive, Chronic kidney disease, Type 2 diabetes mellitus, Colon cancer, Hemicolectomy, Hypertension, Anemia **Relevant Interim AEs:** See below **AE resulting in Death: Pulmonary sepsis** (SAE, Life-threatening, Day 153) On Day 57 [06 May 2020], the patient was hospitalized with Cardiac failure congestive (SAE, Hospitalization/Life-threatening, severe, Day 57-ongoing) and Pneumonia (moderate, Day 57-67). Treatment included metoprolol, furosemide, nitrates, IV antibiotics not otherwise specified, hydralazine, and darbepoetin. On Day 70 he had Atelectasis (SAE, Hospitalization/Life-threatening, severe, Day 70-ongoing with CT scan showing pleural effusion and left lower lobe collapse; treatment included thoracentesis. On Day 121 he was diagnosed with end-stage heart failure and discharged. On Day 153, he was admitted for Pulmonary sepsis with one month progressive decline, and recent decreased level of consciousness, blood pressure, and oxygen saturations. White blood count (WBC) and lactate were elevated; chest radiography had right lower lobe pneumonic consolidation and left-sided pleural effusion. Treatment included ceftriaxone, azithromycin, piperacillin/tazobactam. He died on the same day; autopsy was not performed.

Patient	Brief Vignette
2019-01 44 y.o. White Female RBX2660 Day 1	**Relevant past medical history:** Spina bifida, Hydrocephalus, Ventriculo-peritoneal shunt, Seizure, Paraplegia, Decubitus ulcer, Osteomyelitis **Relevant Interim AEs:** Diarrhea and Fatigue (both - mild, Day29-35). **AE resulting in Death: Spina bifida** (SAE, Death, life-threatening, Day 35-36) On Day 35 the patient was reported to have experienced complications of spina bifida not otherwise specified and was admitted to hospice. On Day 36 she died; cause of death was reported as osteomyelitis of the coccyx and spina bifida, with *C. difficile* colitis and seizures as contributing. **Included as death due to AEs within 6 months of last RBX2660 in error due to imputation as event onset not in database.**