STATISTICAL ANALYSIS PLAN

SPONSOR:

Ironwood Pharmaceuticals, Inc.

100 Summer Street, Suite 2300

Boston, MA 02110

STUDY DRUG:

MD-7246

PROTOCOL NUMBER:

MCP-103-205

STUDY TITLE:

A Phase 2, Randomized, Double-blind, Placebo-controlled, Parallel-group, Dose-range-finding Study of MD-7246 Administered Orally for 12 Weeks to Treat Abdominal Pain in Patients with Diarrhea-predominant Irritable Bowel Syndrome

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LIST OF ABBREVIATIONS

Abbreviation	Term
AE	adverse event
ALT	alanine aminotransferase
ANCOVA	analysis of covariance
APC	Abdominal Pain and Constipation
AST	aspartate aminotransferase
BM	bowel movement
BP	blood pressure
BPM	beats per minute
BSFS	Bristol Stool Form Scale
CBC	complete blood count
CDC HRQOL-4	CDC Healthy Days Core Module
CIC	chronic idiopathic constipation
CMH	Cochran Mantel-Haenszel
CSBM	complete spontaneous bowel movement
eCRF	electronic case report form
eDiary	electronic diary
EOT	End-of-Treatment
HEENT	head, ears, eyes, nose, throat
IBS-D	irritable bowel syndrome with diarrhea
IBS-SSS	Irritable Bowel Syndrome – Symptom Severity Scale
ICF	informed consent form
IPD	Important Protocol Deviations
ITT	Intent-to-treat
IWRS	interactive web response system
LLN	lower limit of normal
LS	least-squares
MedDRA	Medical Dictionary for Regulatory Activities Terminology
MI	multiple imputation
MMRM	Mixed Model Repeated Measures

NRS	numerical rating scale
PCS	potentially clinically significant
PID	patient identification number
Pg	picogram
PGI-C	patient global impression of change
PGI-S	patient global impression of severity
SAE	serious adverse event
SAP	statistical analysis plan
SD	standard deviation
SF-12v2	Short Form-12 Health Survey version 2
SI	*Le Système International d'Unités* (International System of Units)
SOC	system organ class
TEAE	treatment-emergent adverse event
ULN	upper limit of normal
US	United States

1. INTRODUCTION

The MCP-103-205 study is a multicenter, randomized, double-blind, placebo-controlled, parallel-group, dose-range-finding, 12-week study, consisting of 4 distinct periods: up to 2 weeks Screening, 2-3 weeks Pretreatment, 12 weeks Treatment, and 2 weeks Posttreatment. The study will enroll approximately 368 patients who have IBS-D diagnosed using Rome IV criteria. Eligible patients will be randomized in equal proportions to 1 of 4 treatments: 300, 600, or 1200 μg of MD-7246, or matching placebo, administered once daily. MD-7246 is a delayed release (DR) tablet formulation of linaclotide designed to release linaclotide in the distal ileum near the ileocecal junction, to target guanylate cyclase C (GC-C) receptors in the colon and minimize secretory effects in the gastrointestinal (GI) tract.

This statistical analysis plan (SAP) describes the statistical analysis methods for the analysis of efficacy and safety data collected according to the following documents:

- Study Protocol MCP-103-205 Amendment 1, dated 29 January 2019
- Case Report Form (CRF) version 3.0, dated 26 November 2019

Data presentation details for the analyses included in this SAP will be specified in a separate data presentation plan for the format of Tables, Figures and Listings.

2. STUDY OBJECTIVES

The objectives of this study are to evaluate the safety and tolerability, treatment effect on abdominal pain, and dose response of MD-7246 administered orally to patients with diarrhea-predominant irritable bowel syndrome (IBS-D). An exploratory objective of the study is to assess bowel function changes with MD-7246 in patients with IBS-D.

3. STUDY DESIGN

3.1. General Description

This is a multicenter, randomized, double-blind, placebo-controlled, parallel-group, dose-range-finding, 12-week study, consisting of 4 distinct periods, as illustrated in Figure 1 below. The study will enroll patients who have IBS-D diagnosed using Rome IV criteria. Eligible patients will be randomized in equal proportions to 1 of 4 treatments: 300, 600, or 1200 µg of MD-7246, or matching placebo, administered once daily. MD-7246 is a delayed release (DR) tablet formulation of linaclotide designed to release linaclotide in the distal ileum near the ileocecal junction, to target guanylate cyclase C (GC-C) receptors in the colon and minimize secretory effects in the gastrointestinal (GI) tract.

Study Periods

Screening Period: The Screening Period starts with the signing of the informed consent form (ICF; Appendix 1) and may last for up to 28 days. During this period, patient eligibility for entry into the Pretreatment Period will be determined. Loperamide, a protocol-permitted over-the-counter (OTC) medication for diarrhea, will be distributed to eligible patients beginning at the Screening Visit. The end of the Screening Period coincides with the start of the Pretreatment Period. If the patient meets the entry criteria assessed at the Screening Visit and does not require a washout of prohibited medicines, the Screening Visit and Pretreatment Visit may be combined into one visit.

Pretreatment Period: The Pretreatment Period is defined as the 14 to 21 days immediately before the Randomization Visit. During this period, patients will provide the following information in a handheld electronic diary (eDiary):

- Daily Assessments:
  - Daily Abdominal Symptom Assessments in a daily evening report
  - Bowel Movement (BM)-related Assessments on an event-driven basis (meaning these are assessments made for each event at the time the event occurs [or during the daily evening report for any events not previously entered for that day])
- Weekly Assessments:
  - Weekly Patient Assessment of Degree of Relief of IBS Symptoms
  - Weekly Patient Assessment of Adequate Relief of IBS Pain
  - Weekly Patient Assessment of BM-related Symptom Severity (patient global impression of severity [PGI-S])
  - Weekly Patient Assessment of BM-related Symptom Change (patient global impression of change [PGI-C])
- Use of Loperamide for Diarrhea on an event-driven basis

Patients who satisfy all entry criteria will enter the Treatment Period.

Treatment Period: The Treatment Period begins with treatment assignment and lasts for 12 weeks. Patients will be randomized in equal proportions to 1 of 4 treatments: 300, 600, or 1200 µg of MD-7246, or matching placebo. Patients will take their initial dose of study drug at the study center during the Randomization Visit. On all other days, study drug will be taken once daily at approximately the same time of day without regard to food (patients will be instructed to choose a time that is convenient for them and continue daily dosing at that time throughout the Treatment Period). Patients will continue to use the handheld eDiary to provide their:

- Daily Assessments:
  - Daily Abdominal Symptom Assessments in a daily evening report
  - BM-related Assessments on an event-driven basis
- Weekly Assessments:
  - Weekly Patient Assessment of Degree of Relief of IBS Symptoms
  - Weekly Patient Assessment of Adequate Relief of IBS Pain
  - Weekly Patient Assessment of Treatment Satisfaction
  - Weekly Patient Assessment of BM-related Symptom Severity (PGI-S)
  - Weekly Patient Assessment of BM-related Symptom Change (PGI-C)
- Use of Loperamide for Diarrhea on an event-driven basis

Health-related quality-of-life and patient-outcome assessments will be performed at the Randomization Visit and at study visits throughout the Treatment Period. A list of these assessments and the visits when they will be performed is provided in the Schedule of Evaluations.

Patients will complete a Week 2 Phone Call, and Week 4, Week 8, and Week 12/End of Treatment (EOT) Visits during the Treatment Period (see Schedule of Evaluations).

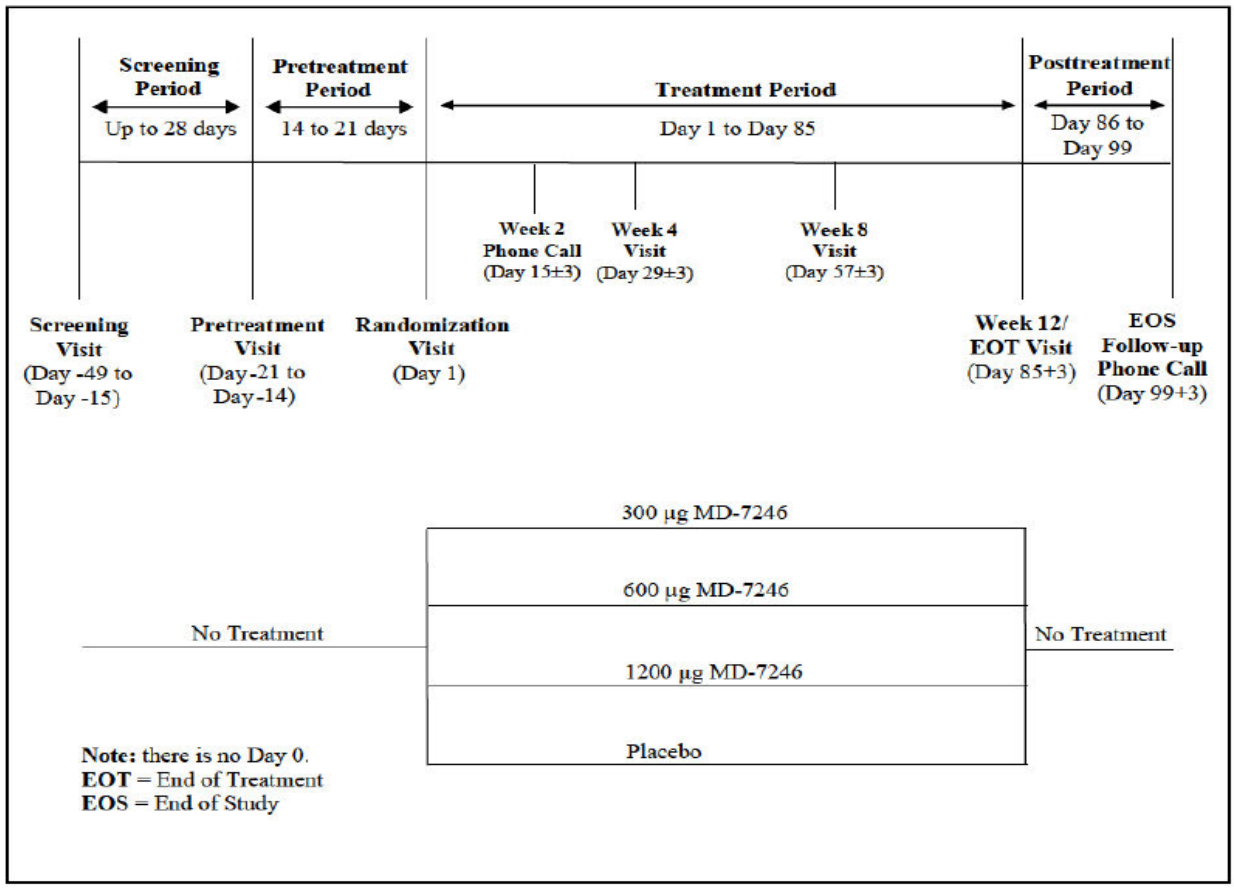
Posttreatment Period: The Posttreatment Period starts on the day following the last day of dosing (Week 12/EOT Visit) and finishes 2 weeks later at the End of Study (EOS) Follow-up Phone Call. During the call, patients will be asked to report any AEs and medicines taken since the Week 12/EOT Visit and detail any other symptoms or comments they may have (at the discretion of the investigator, patients may be requested to return to the study center for their EOS Follow-up).

Overall Study Stopping Criteria and Data Monitoring Committee

The Sponsor may stop enrollment prematurely because of low recruitment rates. The Sponsor may suspend or terminate the study prematurely because of a change in opinion of the Institutional Review Board, Data Monitoring Committee (DMC), or a regulatory authority decision.

An independent DMC will regularly review safety data during the conduct of the study. Periodic safety review meetings will be scheduled when approximately 1/3 and 2/3 of patients have been randomized. The committee will review accumulated safety data, and may request unblinding of the treatment groups, in order to make any recommendations regarding the acceptability of continuing the study based on AEs reported.

Figure 1: Overview of Study Design



3.2. Discussion of Study Design, Including Choice of Control Group

A double-blind, placebo-controlled, parallel-group study design was chosen in accordance with the concepts in ICH E10, Choice of Control Groups and Related Issues in Clinical Trials, in order to provide comparable treatment groups and minimal chance of selection or investigator bias. The study has a 14-21 day Pretreatment Period to establish patient baseline values without study drug and to familiarize patients with data collection methodology (ie, eDiary), a 12-week Treatment Period to compare the test treatment to a placebo control, and a 2-week Posttreatment Period to assess safety after MD-7246 treatment has been withdrawn.

The study will evaluate the effect of the test treatment on abdominal pain in IBS-D patients. Because the test treatment is not expected to significantly affect bowel function, loperamide will be provided beginning at the Screening Visit for patients to use as needed, at the patients' discretion, to stabilize their diarrhea throughout the study; utilization of loperamide for diarrhea will be recorded on an event-driven basis.

Three doses of MD-7246 were selected to assess the relationship between dose and response.

3.3. Treatments Administered

The Treatment Period begins with treatment assignment and lasts for 12 weeks. Patients will be randomized in equal proportions to 1 of 4 treatments: 300, 600, or 1200 µg of MD-7246 or matching placebo. Patients will take their initial dose of study drug at the study center during the Randomization Visit. On all other days, study drug will be taken once daily at approximately the same time of day without regard to food (patients will be instructed to choose a time that is convenient for them and continue daily dosing at that time throughout the Treatment Period).

3.4. Methods of Assigning Subjects to Treatment Groups

Patients who meet all of the inclusion criteria and none of the exclusion criteria will be randomized into the study at the Randomization Visit on Day 1. Approximately 368 patients will be randomized to 1 of 4 treatments: MD-7246 300 µg, 600 µg, or 1200 µg or placebo (1:1:1:1). Randomization numbers will be assigned by interactive web response system (IWRS).

3.5. Blinding

This is a double-blind study. The Sponsor study personnel, the investigator, all other site study personnel, and the patient will remain blinded to individual patient treatment assignments throughout the study. Specifically designated personnel in the Global Patient Safety (GPS) group at Ironwood may be unblinded to the treatment assignment of individual patients for regulatory reporting purposes.

Patient randomization codes for the Treatment Period will be generated by Allergan and implemented by the IWRS vendor (an electronic version will be stored on a secure server). The randomization list will identify each patient by randomization number and include the patient's corresponding treatment assignment. The medication code list will be supplied by [redacted] to the IWRS vendor.

In case of an emergency, the IWRS will be accessed to obtain the study drug assignment of a patient. Accessing the IWRS for emergency unblinding should be done only in an emergency that necessitates identifying the treatment assignment for the welfare of the patient, and only after unsuccessfully attempting to contact the Medical Monitor, or designee. If the blind is broken, the study center will notify the Sponsor immediately. An explanation for breaking the blind will be recorded on the relevant eCRF. Breaking the blind at the study center will disqualify the patient from further participation in the study (refer to Protocol Section 6.3 for details regarding procedures and follow-up for patients who are discontinued from therapy).

4. DETERMINATION OF SAMPLE SIZE

In a previously conducted Phase 2b trial utilizing MD-7246 (ie, Study MCP-103-204), the observed sample standard deviation (SD) for the change from baseline to Week 12 in abdominal pain at its worst ranged from 2.32 to 2.88. Assuming a clinically meaningful treatment difference between MD-7246 and placebo of 1.0 for the mean change from baseline to Week 12 in abdominal pain at its worst, an SD of 2.4, and 92 patients per treatment group (368 patients total), the estimated power is approximately 80% to detect a difference between each MD-7246 dose and placebo at a 2-sided significance level of 0.05 using a 2-sample t-test.

5. PHARMACOKINETICS, EFFICACY, AND SAFETY ASSESSMENTS

5.1. Study Schematic and Schedule of Assessments

The schedule of evaluations for Study MCP-103-205 is presented in Table 1.

Table 1: Schedule of Evaluations

MCP-103-205								
Study Period →	**Screening Period (Up to 28 days)**	**Pretreatment Period (14 to 21 days)**	**Treatment Period (12 weeks)**					**Posttreatment Period (2 weeks)**
Visit Days →	**Screening Visit (Day -49 to Day -15)**	**Pretreatment Visit (Day -21 to Day -14)**	**Random-ization Visit (Day 1)**	**Week 2 Phone Call (Day 15±3)**	**Week 4 Visit (Day 29±3)**	**Week 8 Visit (Day 57±3)**	**Week 12/ End-of-Treatment Visit [p] (Day 85+3)**	**End-of-Study Follow-up Phone Call (Day 99+3)**
Visit Numbers →	**Visit 1**	**Visit 2**	**Visit 3**	**Telephone Call**	**Visit 4**	**Visit 5**	**Visit 6**	**Telephone Call**
Study Procedure ↓								
Signature of ICF	X							
Inclusion and Exclusion Criteria Verification	X	X	X*					
IWRS Registration [a]	X	X	X†		X	X	X	
Medical History	X							
Physical Examination [b]	X						X	
Body Weight and Height [c]	X	X	X*		X	X	X	
Seated Vital Signs [d]	X	X	X*		X	X	X	
Prior and Concomitant Medicines [e]	X	X	X*	X	X	X	X	X
Clinical Laboratory Tests [f]	X		X*		X	X	X	
Pregnancy Test [g]	X		X*				X	
Prohibited Medications Washout Instructions	X							
Loperamide for Diarrhea Dispensed [h]	X	X	X		X	X	X	
AE Evaluations [i]		X	X*	X	X	X	X	X
eDiary Registration and Training [j]		X	X*		X	X	X	
Pain Catastrophizing Scale		X						
Daily and Weekly Assessments [k]		X						
Patient eDiary Entry, in Clinic		X	X*		X	X	X	
eDiary Compliance Verification and Reminder [l]			X*	X	X	X	X	

MCP-103-205								
Study Period →	**Screening Period (Up to 28 days)**	**Pretreatment Period (14 to 21 days)**	**Treatment Period (12 weeks)**					**Posttreatment Period (2 weeks)**
Visit Days →	**Screening Visit (Day -49 to Day -15)**	**Pretreatment Visit (Day -21 to Day -14)**	**Random-ization Visit (Day 1)**	**Week 2 Phone Call (Day 15±3)**	**Week 4 Visit (Day 29±3)**	**Week 8 Visit (Day 57±3)**	**Week 12/ End-of-Treatment Visit [p] (Day 85+3)**	**End-of-Study Follow-up Phone Call (Day 99+3)**
Visit Numbers →	**Visit 1**	**Visit 2**	**Visit 3**	**Telephone Call**	**Visit 4**	**Visit 5**	**Visit 6**	**Telephone Call**
Study Procedure ↓								
Randomization			X†					
SF-12v2			X†		X	X	X	
EQ-5D-3L			X†		X	X	X	
IBS-SSS			X†		X	X	X	
Study Drug Dispensed			X		X	X		
Study Drug Administration [m]			X					
Safety Phone Call [n]				X				X
Study Drug Accountability					X	X	X	
Investigator Consultation with Patient [o]		X (Activity Triggered by eDiary Alert)						

AE=adverse event; BM=bowel movement; CBC=complete blood count; eDiary=electronic diary; EOS=End-of-Study; EOT=End-of-Treatment; EQ-5D-3L=EuroQol-5 Dimension 3-level; IBS=irritable bowel syndrome; IBS-SSS= Irritable Bowel Syndrome-Symptom Severity Scale; ICF=informed consent form; IWRS=interactive web response system; SF-12v2=Short Form-12 Health Survey version 2.

* Assessment done prior to randomization; † Assessment done predose

a. Site personnel will interact with IWRS to register the patient visit. Refer to the IWRS User Manual.
b. A physical examination includes, at minimum, the following: general appearance, HEENT (head, ears, eyes, nose, and throat), cardiac, respiratory, gastrointestinal, musculoskeletal, neurological, and dermatological systems. Rectal examinations should be performed at the discretion of the investigator; the purpose of the rectal examination is to rule out pathologies that might be caused by obstruction. Breast and genitourinary examinations are not required.
c. Height is measured only at the Screening Visit.
d. Vital signs must be obtained in the seated position and include oral temperature, respiratory rate, blood pressure, and pulse. Pulse and blood pressure readings will be taken after the patient has been sitting for 5 minutes.
e. At the Screening Visit, information related to concomitant medicine that patients are taking on the day of the visit will be captured as well as their history of using particular treatments for IBS (approved or unapproved).
f. Chemistry and CBC. Urine drug screen will be performed at the Screening Visit only.

g. Required for females of childbearing potential only. To be eligible to continue in the study, a negative serum pregnancy test must be documented at the Screening Visit. A negative urine pregnancy test must be documented at the Randomization Visit (prior to dosing), and serum pregnancy test results must be documented at the Week 12/EOT Visit.
h. Protocol-permitted loperamide for diarrhea will be supplied to patients at the Screening Visit and, if needed, at the Pretreatment Visit, subsequent study drug dispensing visits, and the Week 12/EOT Visit.
i. All AEs occurring after the patient signs the ICF will be captured.
j. At the Pretreatment Visit and all subsequent visits, the study coordinator will register the patient visit to the clinic in the patient's eDiary. At the Pretreatment Visit, the study coordinator will instruct the patients about the use of the eDiary. Refer to the eDiary User Manual.
k. An eDiary will be used for recording Daily Abdominal Symptom Assessments, BM-related Assessments, Weekly Assessments, Pain Catastrophizing Scale (Pretreatment Visit only), SF-12v2, EQ-5D, IBS-SSS, and any use of loperamide for diarrhea. Patients will enter BMs and loperamide use in the eDiary on an event-driven basis, and will complete an evening entry each day to record daily assessments, including any BMs and/or loperamide use not previously recorded for that day, and weekly assessments. At the Randomization Visit, eDiary assessments will be completed in clinic prior to randomization.
l. At the Randomization Visit and all subsequent visits, the study coordinator will review the eDiary entry information to verify patient compliance with the daily eDiary completion requirement. After determining the patient's compliance, the study coordinator will remind patients to complete the eDiary daily (except at the EOT Visit); eDiary compliance reminder will also occur during the Week 2 Phone Call.
m. Study drug will be administered in the clinic at the Randomization Visit. On all other days, study drug will be taken once daily at approximately the same time of day without regard to food.
n. Appropriate site personnel will contact patients by phone to assess for AEs, record concomitant medicines, and (at Week 2 only) remind patients to comply with study drug and eDiary completion. Any AE or concomitant medicine reported during the phone call will be captured on the eCRF. Patients who report AEs during the call will be provided with instructions on the appropriate follow-up care. At the discretion of the investigator, patients may be requested to return to the clinic for this contact or an unscheduled visit may be conducted following the phone call.
o. Throughout the Pretreatment and Treatment Periods, the eDiary will send automated alerts to the investigator if patients experience episodes of constipation with excessive use of loperamide or diarrhea that is not well managed with the use of loperamide (as defined in Protocol Section 8.2.6). In the event of an alert, the investigator must review with the patient their use of loperamide and their pattern of constipation or diarrhea.
p. Patients who are randomized but do not complete the Treatment Period (withdraw consent or are discontinued before they have completed 12 weeks of treatment), will be considered Treatment Period withdrawals and should complete the procedures required at the EOT Visit (even if out of window).

5.2. Pharmacokinetic Assessments

There are no pharmacokinetic assessments planned for this study.

5.3. Efficacy Assessments

During the Pretreatment and Treatment Periods, patients will enter information into the eDiary. Certain information will be entered by the patient on an event-driven basis, in a daily evening report, and on a weekly basis, as specified below. Event-driven and daily assessments of abdominal and bowel symptoms will be collected using the Diary of IBS Symptoms – Diarrhea (DIBSS-D; Protocol Appendix 4).

5.3.1. Key Efficacy Assessment

The efficacy assessment that will be used to determine the key efficacy endpoints is the daily patient assessment of abdominal pain at its worst.

Daily Patient Assessment of Abdominal Pain at its Worst

Patient assessment of abdominal pain at its worst will be collected via a daily evening report in the eDiary. The rating of abdominal pain at its worst during the previous 24 hours on an 11-point numerical rating scale (NRS) will be provided by the patient answering the following question:

"How would you rate your worst abdominal pain in the past 24 hours?"

<eDiary presents NRS where 0 is anchored with "No abdominal pain" and 10 is anchored with "Worst possible abdominal pain">

5.3.2. Additional Efficacy Assessments

Daily Patient Assessment of Abdominal Bloating at its Worst

Patient assessment of abdominal bloating at its worst will be collected via a daily evening report in the eDiary. The rating of abdominal bloating at its worst during the previous 24 hours on an 11-point NRS will be provided by the patient answering the following question:

"How would you rate your worst abdominal bloating in the past 24 hours?"

<eDiary presents NRS where 0 is anchored with "No abdominal bloating" and 10 is anchored with "Worst possible abdominal bloating">

Daily Patient Assessment of Abdominal Discomfort at its Worst

Patient assessment of abdominal discomfort at its worst will be collected via a daily evening report in the eDiary. The rating of abdominal discomfort at its worst during the previous 24 hours on an 11-point NRS will be provided by the patient answering the following question:

"How would you rate your worst abdominal discomfort in the past 24 hours?"

<eDiary presents NRS where 0 is anchored with "No abdominal discomfort" and 10 is anchored with "Worst possible abdominal discomfort">

Daily Patient Assessment of Abdominal Cramping at its Worst

Patient assessment of abdominal cramping at its worst will be collected via a daily evening report in the eDiary. The rating of abdominal cramping at its worst during the previous 24 hours on an 11-point NRS will be provided by the patient answering the following question:

"How would you rate your worst abdominal cramping in the past 24 hours?"

<eDiary presents NRS where 0 is anchored with "No abdominal cramping" and 10 is anchored with "Worst possible abdominal cramping">

Weekly Patient Assessment of Degree of Relief of IBS Symptoms

Patient assessment of degree of relief of IBS symptoms will be reported weekly by eDiary entry. The rating of degree of relief during the previous 7 days on a 7-point balanced scale will be provided by the patient answering the following question:

"Compared to before you started the study, how would you rate your IBS symptoms during the past 7 days?"

1=Significantly relieved

2=Moderately relieved

3=Somewhat relieved

4=Unchanged

5=Somewhat worse

6=Moderately worse

7=Significantly worse

Weekly Patient Assessment of Adequate Relief of IBS Pain

Patient assessment of adequate relief of IBS pain will be reported weekly by eDiary entry. The rating of adequate relief during the previous 7 days on a binary scale will be provided by the patient answering the following question:

"Overall, have you had adequate relief from your IBS pain during the past 7 days?"

1=Yes

2=No

Weekly Patient Assessment of Treatment Satisfaction

Patient assessment of treatment satisfaction will be reported weekly by eDiary entry each week of the Treatment Period after the Randomization Visit. Patients will answer the following question on a 5-point ordinal scale:

"Overall, how satisfied are you with the study medication's ability to relieve your IBS symptoms?"

1=Not at all satisfied

2=A little satisfied

3=Moderately satisfied

4=Quite satisfied

5=Very satisfied

5.3.3. Exploratory Assessments

In addition to the key/additional efficacy assessments, the following non-efficacy exploratory assessments are used in determining the exploratory endpoints.

Each day of the Pretreatment and Treatment Periods, the patient will complete eDiary entries on an event-driven basis to report BMs. The patient is also asked to provide assessments of stool consistency and urgency, which are described below. Additionally, the patient will complete eDiary entries on an event-driven basis to report use of loperamide for diarrhea. Patients will complete a daily evening report to enter any BMs and/or loperamide usage not previously reported by the patient for that day (recall is limited to the period of time following the patient's completion of the previous night's Evening Report [ie for an Evening Report completed yesterday at 10:00pm, no events can be recorded today with a time preceding 10:00pm yesterday], or time following 7:00pm the previous day in instances where the previous night's Evening Report was missed by the patient).

Bowel Movement Frequency

The assessment of BM frequency is based on the eDiary questions that record BM date and time. Patients will report BMs on an event-driven basis. An evening report will ask whether the patient entered all BMs for that day, and collect BMs not previously entered.

Stool Consistency (5-point ordinal scale)

Patient assessment of stool consistency will be collected using two eDiary questions recorded on an event-driven basis (ie, for each BM reported in the eDiary). For each BM, patients will assess stool consistency by daily eDiary entry using a 5-point ordinal scale:

"How would you describe the consistency of your stool?"

1=Very hard

2=Hard

3=Neither too hard nor too soft

4=Loose but not watery

5=Very loose and watery

The patient also assesses his/her stool consistency using the 7-point Bristol Stool Form Scale (BSFS) described below.

Stool Consistency (BSFS)

In addition to the patient assessment of stool consistency using the 5-point ordinal scale (described above), the stool consistency of each BM will be recorded by the patient by daily eDiary entry for each BM on an event-driven basis using the BSFS (Protocol Appendix 5), which depicts the stool consistency characteristics along with descriptions for each of them. The patient assigns a corresponding score for each BM. The BSFS is a well-accepted and widely-used measurement of stool consistency. The 7-point ordinal BSFS is provided below (see Protocol Appendix 5 for full scale including pictures):

"Please describe the form of your stool using the following scale where:"

1=Separate hard lumps like nuts (hard to pass)

2=Sausage-shaped but lumpy

3=Like a sausage but with cracks on the surface

4=Like a sausage or snake, smooth and soft

5=Soft blobs with clear-cut edges (passed easily)

6=Fluffy pieces with ragged edges, a mushy stool

7=Watery, no solid pieces, entirely liquid

BM Urgency

Patient assessment of BM urgency will be collected by eDiary entry on an event-driven basis (ie, for each BM reported in the eDiary). For each BM, urgency will be assessed by the patient using a binary scale:

"Did you feel the need to rush to the toilet in order to avoid an accident before your bowel movement?"

1=Yes

2=No

Use of Loperamide for Diarrhea

Patients will report use of loperamide (Imodium®) for diarrhea on an event-driven basis (ie, for each use of the medication). An evening report will ask whether the patient entered all loperamide use for that day and collect all loperamide use not previously entered for that day (recall is limited to the period of time following the patient's completion of the previous night's Evening Report [ie for an Evening Report completed yesterday at 10:00pm, no events can be recorded today with a time preceding 10:00pm yesterday], or time following 7:00pm the previous day in instances where the previous night's Evening Report was missed by the patient).

"Did you use Imodium® (loperamide) for diarrhea that you have not yet reported in the Diary?"

1=Yes

2=No

"Was the Imodium® (loperamide) taken yesterday or today?"

1=Yesterday

2=Today

"How many Imodium® (loperamide) pills did you take?"

__ pill(s)

Weekly Patient Assessment of Bowel Movement-related Symptom Severity

Patient global impression of severity (PGI-S) of BM-related symptoms will be reported weekly by eDiary entry. The rating of BM-related symptom severity during the previous 7 days on a 5-point ordinal scale will be provided by the patient answering the following question:

"How would you rate your bowel movement-related symptoms during the past 7 days?"

1=None

2=Mild

3=Moderate

4=Severe

5=Very severe

Weekly Patient Assessment of Bowel Movement-related Symptom Change

Patient global impression of change (PGI-C) of BM-related symptoms will be reported weekly by eDiary entry. The rating of BM-related symptom change during the previous 7 days on a 7-point ordinal scale will be provided by the patient answering the following question:

"Compared to before you started the study, please rate the overall change in your bowel movement-related symptoms during the past 7 days."

1=Very much better

2=Moderately better

3=A little better

4=No change

5=A little worse

6=Moderately worse

7=Very much worse

5.4. Health Outcomes Assessments

Pain Catastrophizing Scale

The Pain Catastrophizing Scale is an instrument for assessing catastrophic thinking related to pain and consists of 13 questions assessing the following 3 dimensions: rumination, magnification, and helplessness (Protocol Appendix 6). The Pain Catastrophizing Scale will be completed at the Pretreatment Visit as specified in the Schedule of Evaluations; the assessment

will be triggered by site personnel on the patient's eDiary for the questionnaire to be self-administered by the patient.

Short Form-12 Health Survey version 2 (SF-12v2)

The SF-12v2 is a widely used generic measure of health status (Protocol Appendix 7). The SF-12v2 measures 8 concepts commonly represented in widely used surveys: physical functioning, role limitations due to physical health problems, bodily pain, general health, vitality (energy/fatigue), social functioning, role limitations due to emotional problems, and mental health (psychological distress and psychological well-being). These 8 scales are aggregated into 2 summary measures: the Physical and Mental Component Summary Scores. The SF-12v2 will be completed at the Randomization Visit prior to the patient receiving study drug and at subsequent visits as specified in the Schedule of Evaluations. At each of the designated visits, the assessment will be triggered by site personnel on the patient's eDiary for the questionnaire to be self-administered by the patient.

EuroQol-5 Dimension 3-level (EQ-5D-3L)

The EQ-5D-3L is a generic measure of health status that is widely used in Europe (protocol Appendix 8). The descriptive system consists of 5 questions assessing the following dimensions: mobility, self-care, usual activities, pain/discomfort, and anxiety/depression. Responses to the 5 questions define a health state for which a utility index can be derived from published algorithms. The second component of the EQ-5D-3L is a visual analogue scale, asking patients to rate their health from 0 to 100 (0 represents worst imaginable health state and 100 represents best imaginable health state). The EQ-5D-3L will be completed at the Randomization Visit prior to the patient receiving study drug and at subsequent visits as specified in the Schedule of Evaluations. At each of the designated visits, the assessment will be triggered by site personnel on the patient's eDiary for the questionnaire to be self-administered by the patient.

Irritable Bowel Syndrome-Symptom Severity Scale (IBS-SSS)

The IBS-SSS contains 7 questions that ask patients to rate the severity and frequency of their abdominal pain and distention, dissatisfaction with bowel habits, and the extent to which the patient's bowel habits interfere with life in general (Protocol Appendix 9). The IBS-SSS will be completed at the Randomization Visit prior to the patient receiving study drug and at subsequent visits as specified in the Schedule of Evaluations. At each of the designated visits, the assessment will be triggered by site personnel on the patient's eDiary for the questionnaire to be self-administered by the patient.

5.5. Completion of eDiary Assessments

To ensure patient compliance with the completion of the evening eDiary report, alarms occurring at 7, 8, 9, 10, and 11 PM will be programmed to alert the patient that he/she has not completed the evening eDiary report. Upon completion of the evening eDiary report, the alarm will no longer be active for the remainder of that day. If the patient does not complete the evening report prior to midnight, that day's evening report will no longer be available for reporting by the patient, and all evening assessments for that day will be considered missing.

Patient compliance with the daily eDiary completion requirement will be verified by site staff as specified in the Schedule of Evaluations.

5.6. Safety Assessments

Safety will be evaluated by AE reports (discussed herein), standard clinical laboratory assessments, vital signs, physical examinations, and medical history. Planned timepoints for all safety assessments are provided in the Schedule of Evaluations.

5.6.1. Adverse Events

Adverse Event

An AE is any untoward medical occurrence associated with the use of a drug in humans, whether or not considered drug-related.

An AE, therefore, can be any unfavorable and unintended sign (including an abnormal laboratory finding, for example), symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Note: A procedure is not an AE, but the reason for a procedure may be an AE.

Treatment-emergent Adverse Event

A TEAE is an event that emerges, or a preexisting event that worsens (with a new start date), any time after administration of the first dose of study drug (Day 1) during the treatment emergent period, which is defined as the date of the first treatment to the date on or before 1 day after the date of the last treatment. The Investigator will provide an assessment of the AE relationship to study drug and AE severity for each AE.

5.6.1.1. Causality Assessment

For all AEs, the investigator must provide an assessment of causal relationship to study drug. The investigator must assess the relationship of each AE (including SAEs) to the use of a study drug using a 2-category scale (not related or related) based on clinical judgment and using all available information, and may include consideration of the following factors:

- Possible alternative causes of the AE, including the disease under treatment, preexisting conditions, concomitant use of other drugs, and presence of environmental or genetic factors
- The temporal association between drug exposure and onset of the AE
- Whether the manifestations of the AE are consistent with known actions or toxicity of the investigational product
- Whether the AE resolved or improved with stopping use of the study drug; judgment should be used if multiple products are discontinued at the same time

The causality assessment must be recorded in the patient's source documentation and on the AE page of the patient's eCRF. The causal relationship between study drug and the AE will be assessed using the categories shown in the following table:

Category	Definition
Not related	An AE is not associated with study medication if: - Lack of a temporal relationship to study drug administration makes a causal relationship improbable (eg, the event did not occur within a reasonable time frame following administration of the study medication); and/or - Other causative factor(s) (eg, a preexisting clinical condition, other concomitant treatments) more likely explain the occurrence of the event, and - The event did not improve with stopping of the investigational product, and/or - The event did not recur upon re-exposure with investigational product
Related	An AE is attributed to the study medication if: - A temporal relationship to study drug administration makes a causal relationship plausible (eg, the event occurred within a reasonable time frame following administration of study medication); and/or - Other causative factor(s) (eg, the patient's clinical condition, other concomitant treatments) either do not explain the event or are less equally likely to have led to the occurrence of the event, or - The event improved with stopping of the investigational product, and/or - The event recurred upon re-exposure with investigational product

5.6.1.2. Classification of Adverse Event Severity

The investigator or delegated physician will provide an assessment of the severity of each AE by recording a severity rating in the patient's source documentation and on the AE page of the patient's eCRF. *Severity*, which is a description of the intensity of manifestation of the AE, is distinct from *seriousness*, which implies a patient outcome or AE-required treatment measure associated with a threat to life or functionality. Severity will be assessed according to the following scale:

- **Mild:** A type of AE that is usually transient and may require only minimal treatment or therapeutic intervention. The event does not generally interfere with usual activities of daily living.

- **Moderate:** A type of AE that is usually alleviated with additional specific therapeutic intervention. The event interferes with usual activities of daily living, causing discomfort, but poses no significant or permanent risk of harm to the research participant.
- **Severe:** A type of AE that interrupts usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention.

AEs should be recorded using the maximum intensity of the event (eg, if a patient reported nausea lasting 3 days, 1 start date and 1 stop date should be recorded along with the maximum intensity experienced for that event over that 3-day timeframe).

5.6.1.3. Serious Adverse Events

An AE is considered "serious" if, in the view of either the investigator or Sponsor, it results in any of the following outcomes:

- Death
- Life-threatening experience: An AE is considered "life-threatening" if, in the view of either the investigator or Sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an AE that, had it occurred in a more severe form, might have caused death.
- Inpatient hospitalization or prolongation of an existing hospitalization: AEs requiring hospital admissions that are less than 24 hours in duration do not meet this criterion. A scheduled hospitalization for a preexisting condition that has not worsened during participation in the study does not meet this criterion. Preplanned hospitalizations for an elective medical/surgical procedure or routine check-ups do not meet this criterion.
- Persistent or significant disability or incapacity or substantial disruption of the ability to conduct normal life functions
- A congenital anomaly or birth defect
- Is considered to be an important medical event: Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

5.6.2. Clinical Laboratory Data

See below for the list of clinical laboratory tests to be performed and refer to the Schedule of Evaluations for the timing and frequency. All protocol-required laboratory assessments, as defined below, must be conducted in accordance with the laboratory manual and the Schedule of Evaluations.

The tests detailed below will be performed (additional tests may be performed at any time during the study as determined necessary by the investigator or as required by local regulations):

- **Hematology:** Absolute and differential white blood cell count, erythrocyte count, hemoglobin, hematocrit, platelet count, and red blood cell indices (mean corpuscular volume, mean corpuscular hemoglobin, and mean corpuscular hemoglobin concentration)
- **Chemistry:** Sodium, magnesium, potassium, calcium, chloride, glucose, blood urea nitrogen, creatinine, total protein, alkaline phosphatase, albumin, total bilirubin, aspartate aminotransferase, alanine aminotransferase, bicarbonate, phosphate, cholesterol, and uric acid
- **Other:** Urine drug screening (cocaine, barbiturates, amphetamines, opiates, benzodiazepine, alcohol, and cannabinoids) (Screening Visit only). By definition, a positive drug screen (including testing positive for marijuana in states where marijuana is legalized) is clinically significant and, therefore, exclusionary.
- **Pregnancy test:** Serum human chorionic gonadotropin pregnancy test (for women of childbearing potential only) should be conducted at the Screening Visit and Week 12/EOT Visit. A negative urine pregnancy test must be documented at the Randomization Visit for women of childbearing potential to be eligible for randomization and dosing with study drug. Positive results on the pregnancy test will exclude a patient from participating or continuing in the study.

The investigator must review each laboratory report, document this review, and record any clinically relevant changes occurring during the study in the AE section of the eCRF. The laboratory reports must be filed with the source documents. All laboratory tests with abnormal values considered clinically significant during participation in the study should be repeated until the values return to normal or baseline or are no longer considered clinically significant by the investigator or Medical Monitor. If such values do not return to normal/baseline within a period of time judged reasonable by the investigator, the etiology should be identified and the Sponsor notified. If laboratory values from non-protocol specified laboratory assessments performed at the institution's local laboratory require a change in patient management or are considered clinically significant by the investigator (eg, SAE or AE or study drug discontinuation), the results must be recorded in the eCRF.

5.6.3. Vital Signs

Vital sign measurements will be performed as outlined in the Schedule of Evaluations. Oral temperature, pulse rate, respiratory rate, and systolic and diastolic blood pressure (BP) will be assessed with the patient in a seated position. BP and pulse measurements will be assessed with a completely automated device; manual techniques will be used only if an automated device is not available.

BP and pulse measurements should be preceded by at least 5 minutes of rest for the patient in a quiet setting without distractions (eg, television, cell phones).

5.6.4. Physical Examination

A complete physical examination will be performed as outlined in the Schedule of Evaluations, by the investigator or a licensed health professional listed on Form FDA 1572. Any physical examination abnormality that the investigator considers to be potentially clinically significant and changed from the baseline will be reported as an AE. A complete physical examination will include, at a minimum, assessment of the general appearance of the patient and the HEENT (head, eyes, ears, nose, and throat), cardiac, respiratory, gastrointestinal, musculoskeletal, neurological, and dermatological systems. Height (only at screening) and weight will also be measured and recorded.

Rectal examinations should be performed at the discretion of the investigator; the purpose of the rectal examination is to rule out pathologies that might be caused by obstruction. For all physical examinations, the breast and genitourinary examinations are not required.

5.6.5. Medical History

A complete medical history, including disease-specific history, will be provided by the patient at the Screening Visit. As part of their disease-specific history, patients will be asked about any prior IBS diagnosis. Patients will be enrolled based on the Rome IV criteria for IBS-D, as detailed in protocol Section 6.1 and described below.

Patient reports recurrent abdominal pain, on average at least 1 day/week during the 3 months before the diagnosis, with the onset at least 6 months before the diagnosis, associated with 2 or more of the following features:

a. Related to defecation

b. Associated with a change in frequency of stool

c. Associated with a change in form (appearance) of stool

According to the Rome IV criteria, patients are subtyped as IBS-D if they have >25% of BMs with BSFS score of 6 or 7 and <25% of BMs with BSFS score of 1 or 2, based on stool form on days with at least 1 abnormal BM.

6. DATA QUALITY ASSURANCE

6.1. Data Monitoring

Before any patient enters the study, a representative of Ironwood or its authorized designee will meet with the investigator and his/her staff to review the procedures to be followed while conducting the study and to train them on recording the data on the eCRFs using the electronic data capture (EDC) system. After the first patient signs the ICF, the Ironwood representative, a site monitor, will periodically monitor the progress of the study by conducting monitoring visits. This site monitor will also be able to review the status of data queries remotely, possibly warranting more frequent communication with the investigator and his/her staff. The investigator and his/her staff will be responsible for reviewing eCRFs, resolving data queries generated by the site monitor via the system, providing missing or corrected data, approving all changes performed on his/her data, and endorsing the patient data within the EDC system. This approval method will include applying an electronic signature, which is a uniquely assigned username and password that together will represent a traditional handwritten signature.

6.2. Data Recording and Documentation

All data collected in the context of this study will be stored and evaluated in such a way as to guarantee patient confidentiality in accordance with the legal stipulations applying to confidentiality of data. Study records (eg, essential documents [commonly called regulatory documents], correspondence) will be retained at the study center, along with adequate source documentation, according to FDA and ICH requirements. All study records must be available for inspection by Ironwood, its authorized designee, and the FDA.

Data collection will involve the use of the [redacted], to which only authorized personnel will have access. In addition to periodic monitoring occurring within the system by Sponsor site monitors, programmatic edit checks and manual review procedures will be used to review the data for completeness, logic, and adherence to the study protocol. As a result of this monitoring and these checks, data queries may be electronically issued to the clinical study centers and electronically closed by those centers. The identifying information (assigned username, date, and time) for both the originator of the query (if created during the monitoring process) and the originator of the data change (if applicable), as well as the investigator's approval of all changes performed on his/her patients' data, will be collected.

7. STATISTICAL METHODS

7.1. General Methods

In general, efficacy analyses will be performed for all efficacy outcomes reported during the Treatment Period based on the treatment group assigned at the randomization visit. Unless otherwise specified, all confidence intervals (CIs) will be 2-sided and with a confidence level of 95%. No adjustments will be made for multiplicity in the conduct of comparisons among the MD-7246 doses relative to placebo for all efficacy endpoints, unless otherwise specified. Safety analyses will be performed using descriptive summaries based on the Safety Population and on actual treatment received.

For analysis of continuous parameters (eg, change from baseline), descriptive statistics (n, mean, standard deviation [SD], median, and range) will be calculated and presented for each treatment group. For categorical parameters (eg, responder vs. non-responder), the number and percentage of each category will be calculated and presented for each treatment group. Percentages will be based on the total number of non-missing values; the number missing will be presented, but without a percentage.

All statistical analyses will be performed using SAS Version 9.4 (or later) for Windows.

7.2. Adjusting for Covariates

As a general approach, the baseline value will be included in the mixed model for repeated measures (MMRM), Poisson regression model, and/or analysis of covariance (ANCOVA) as a covariate when the postbaseline or change from baseline are analyzed.

7.3. Handling of Dropouts/Missing Data Imputation Method

For weekly responder endpoints based on daily eDiary entries (eg, Weekly Abdominal Pain 30% Responder), a patient who reported daily assess for fewer than 4 days during a week within the Treatment Period will be considered a non-responder for that week. A patient will be deemed a weekly non-responder for all weeks following the patient's discontinuation from the study treatment.

Similarly, the weekly symptom score that is calculated using daily eDiary symptom assessments will be the average of available daily assessment records for at least 4/7 records during the relevant week. The weekly symptom score will be set to missing for weeks with fewer than 4 days of daily assessments. For patients who prematurely discontinues the treatment prior to Week 12, the weekly symptom scores will be considered missing for all weeks after the treatment discontinuation.

Unless stated otherwise, missing data will not be imputed.

7.4. Interim Analysis and Data Monitoring

No interim analysis of unblinded efficacy data will be performed. A DMC will be reviewing the safety data at defined time points (see DMC Charter). During the process, the data review will be handled by an independent clinical research organization (CRO). No sponsors' team members will be exposed to any data in unblinded fashion.

7.5. Multicenter Studies

This study is being conducted in approximately 80 centers in the US. Due to the potential of small numbers of patients per center, study centers may be pooled into the following geographical regions as necessary for the purposes of subgrouping: Northeast, Southeast, Midwest, Southwest, and West, for the relevant analyses such as the summary of disposition.

7.6. Multiple Comparisons/Multiplicity

No multiplicity adjustments will be used.

7.7. Examination of Subgroups

No subgroup analysis is planned; however, specific subgroup analyses may be performed for selected efficacy parameters if data warrants.

7.8. Analysis Methods

7.8.1. Patient Populations

Screened Population

The Screened Population consists of all patients who had a Screening Visit (Visit 1) and were assigned a patient identification (PID) number.

Safety Population

The Safety Population consists of all patients who received at least one dose of study drug. All safety analyses will be based on Safety Population, unless otherwise specified. Patients in Safety Population will be grouped based on the first treatment they receive.

Modified Intent-to-treat Population

The Intent-to-Treat (ITT) Population consists of all randomized patients. The modified ITT population (mITT) consists of all patients within ITT and have at least 1 dose of study drug. All efficacy analyses will be performed on the mITT Population for all outcomes reported during the Treatment Period using treatment group assigned at randomization, unless otherwise specified.

7.8.1.1. Analysis Periods

For the purpose of analyses, three analysis periods are defined and will be used for different analyses.

The Pretreatment period is defined as the 14 days immediately prior to the date of the first dose of study drug.

The Treatment Period is defined as the period from the date of the first dose of study drug to the date of the last dose of study drug. This will be used as the primary efficacy analysis period unless otherwise specified.

The Treatment Emergent Period (TE Period) is defined as the period from the date of the first dose of study drug to 1 day after the date of the last dose. For analysis purposes, AEs and required medications will be considered treatment emergent if they occur within this period. TE period will be the primary analysis period for AEs and medications.

7.8.2. Patient Disposition

The number and percentage of patients included in the Safety and mITT populations will be summarized overall and by treatment group for each trial center within each geographic region. The number of patients in the Screened Population will be summarized by geographic region (See Section 7.5 for definition of geographic region) and within each region by study center.

The number and percentage of screen failure patients (ie, patients who entered the Screening Period but not the Pretreatment Period) and the number and percentage of pretreatment failures (i.e., patients who entered the Pretreatment Period but were not randomized), along with the associated reasons for failure, will be tabulated overall for the Screened Population. Patients who initially failed screening and were re-screened will only be counted once; that is, the PID number used for the rescreened subject will be summarized and listed for patient disposition.

The number and percentage of mITT patients, the number and percentage of patients who completed the Treatment Period, as well as the number and percentage of patients who prematurely discontinued treatment will be presented for each treatment group and overall. The reason for premature discontinuation from treatment as recorded on the trial completion forms of the electronic case report forms (eCRFs) will also be presented. All patients who prematurely discontinue treatment will be listed by discontinuation reason for the ITT Population.

Similarly, the numbers and percentages of patients who completed study (ie, completed the Treatment Period and 2-week Posttreatment Period), and who prematurely discontinue from the study will be summarized with reasons for discontinuation by treatment as recorded on the study completion form.

7.8.3. Protocol Deviations

Protocol deviations and Important Protocol Deviations (IPD) will be identified and documented by the study team for all mITT patients prior to unblinding. IPDs will be determined based on blinded review of all protocol deviations and protocol deviation categories. IPD categories include, but are not limited to:

- Those who entered the study even though they did not satisfy the entry criteria
- Those who developed withdrawal criteria during the study but were not withdrawn
- Those who received the wrong treatment or incorrect dose
- Those who received an excluded concomitant treatment

The number and percentage of subjects with IPDs will be presented by study treatment and IPD category. All protocol deviations regardless of significance will be listed.

A separate summary and listing of all inclusion/exclusion criteria deviations will also be provided. This summary will be based on data as recorded on the inclusion/exclusion page of the eCRF.

7.8.4. Demographics and Other Baselines Characteristics

Demographic parameters (age, age group, race, ethnicity, sex), and baseline characteristics (weight; height; and body mass index, calculated as weight in kg/[height in m]^2) will be summarized descriptively for the Safety and mITT Populations. Baseline efficacy and bowel

parameters (including abdominal pain, abdominal bloating, abdominal discomfort, abdominal cramping, BM frequency, urgent BM frequency, BSFS score, stool consistency) will be summarized descriptively for the mITT Population.

7.8.5. Measurement of Treatment Compliance

Dosing compliance for a specified period is defined as the total number of tablets taken by a patient during that period divided by the number of tablets that were expected to be taken during the same period, multiplied by 100. The total number of tablets taken will be calculated based on the following equation: total number of tablets dispensed – (number of tablets returned + number of tablets lost). The total number of tablets expected to be taken during a specified period equals the number of days in the period, multiplied by 4 (ie, the daily number of tablets to be taken).

Descriptive statistics for study drug compliance for the Treatment Period overall, and for each of the 3 consecutive 4-week periods in the Treatment Period (consistent with study drug dispensing) will be presented for the Safety Population.

7.8.6. Extent of Exposure

Duration of exposure to study drug, calculated as the number of days from the date of first dose taken to the date of last dose taken, inclusive, regardless of the detailed dosing information for individual days in between, will be summarized by treatment group for the Safety Population. Summary statistics including n, mean, standard deviation, median, minimum and maximum will be tabulated. The numbers and percentages of patients completing certain duration periods will also be included for the following: ≥1 week; ≥4 weeks; ≥8 weeks; 12 weeks (including more than 12 weeks).

7.8.7. Prior and Concomitant Medication

Prior and concomitant medicines will be classified using WHO Drug Dictionary version B3 March 2019. Prior medicines are defined as any medicines with a start date prior to the date of first dose of study drug. Concomitant medicines are defined as any medicines with an end date on or after the date of first dose of study drug during the TE Period. Posttreatment medicines are defined as any medicines started after the TE Period (ie, >24h after the last dose of study drug). A medication started prior to the first dose of the study drug and ended on or after the date of the first dose of the study drug will be considered as both prior and concomitant medications.

Both prior and concomitant medicine use, classified by therapeutic class, will be summarized by treatment group and by Anatomical-Therapeutic-Chemical (ATC) classification for the Safety Population. Multiple medicines used by a patient in the same ATC category will be counted only once.

Posttreatment medicines will be presented in data listings only.

7.8.8. eDiary Compliance

eDiary compliance will be based on the percentage of complete daily eDiary entries made by a patient and analyzed base on mITT population. A complete daily eDiary entry is defined as one in which the patient responds to every eDiary question asked in the evening report on a day (namely, a complete eDiary entry day). The questions that are asked weekly will not be included

in the determination of a complete entry. The percentage of complete eDiary entries will be calculated as 100 times the number of completed evening reports divided by the total number of days in the study period (respectively for Pretreatment Period and Treatment Period).

Descriptive statistics for eDiary compliance (%) will be summarized during Pretreatment and Treatment Period, at each week and overall. The patients with ≥80%/<80% complete eDiary entries during the Pretreatment and Treatment Periods will be tabulated by treatment. Numbers and percentages of patients completing at least 4 eDiary evening reports will be summarize at each week during Treatment Period and Pretreatment Period.

7.8.9. Efficacy Endpoints and Analyses

All efficacy analyses will be based on the mITT Population, unless otherwise specified. All hypothesis testing will be performed pairwise without adjustment for multiplicity, unless otherwise specified.

Baseline values for efficacy and exploratory parameters are derived from the eDiary and eCRF data collected in the Pretreatment Period, specifically, the period of time from 14 days prior to Randomization Visit and up to the time of the first dose of study drug.

- For patient symptom parameters (eg, abdominal symptoms [pain, bloating, discomfort, cramping]), the baseline value will be the average of the non-missing severity scores reported during the 14-day prior to the first dose during Pretreatment period and up to time of first dose of study drug.
- The baseline BM and urgent BM weekly rates will be derived based on the number of BMs and urgent BMs a patient had during the 14-day prior to the first dose in Pretreatment Period and up to time of first dose of study drug.
- Baseline BSFS score and stool consistency will be calculated as the average of the non-missing values reported by the patient during the 14-day prior to the first dose in Pretreatment Period and up to time of first dose of study drug.

7.8.9.1. Key Efficacy Endpoints

Change from Baseline in Abdominal Pain Through the Treatment Period

The Change from Baseline in Abdominal Pain Through the Treatment Period is defined as the overall change from baseline in the Abdominal Pain through the Treatment Period, which is estimated based on each weekly change from baseline in the abdominal pain score (based on the daily abdominal pain at its worst score) minus the baseline abdominal pain score for patients in the mITT population.

The postbaseline weekly abdominal pain score is the average of the non-missing daily abdominal pain at its worst scores during a week (Weeks 1-12) if there are at least 4 daily scores entered into the eDairy during the week, or will be considered as missing if there are less than 4 daily scores in the week. Weekly change from baseline will be calculated for each week as the weekly score minus the baseline score.

6/12 Week Abdominal Pain 30% Responder

The 6/12 Week Abdominal Pain 30% Responder is defined for patients in the mITT population and is a patient who meets the Weekly Abdominal Pain 30% Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. A Weekly Abdominal Pain 30% Responder is a patient who has a decrease from baseline of at least 30% in the weekly abdominal pain for that week. If a patient did not have an abdominal pain score for at least 4 days in a particular week during the Treatment Period, the patient will not be considered a Weekly Abdominal Pain 30% Responder for that week. For all weeks after the treatment discontinuation, the patient will be considered as non-responder. A patient in mITT population who did not have any post baseline assessment for abdominal pain at its worst will be considered a weekly non-responder for all post baseline weeks, subsequently, will not be a 6/12 week abdominal pain responder.

7.8.9.2. Analyses of the Key Efficacy Endpoints

Change from Baseline in Abdominal Pain Through the Treatment Period

The overall treatment effect will be evaluated via the comparison between the MD-7246 dose groups and placebo with regard to the weekly change from baseline (CFB) in abdominal pain using a MMRM model, including the weekly CFB score in abdominal pain as dependent variable and treatment, visit week (categorical), and treatment-by-week as fixed effects, patient as a random effect, and the baseline value as a continuous covariate. An unstructured covariance matrix will be used for the MMRM analysis, unless the model does not converge, in which case the compound symmetry will be used.

In this MMRM, a patient in mITT population without any postbaseline abdominal pain score at its worse will be considered missing. For patients with postbaseline abdominal pain scores, at any particular week, if there are fewer than 4 daily scores available, the weekly change from baseline will be considered as missing. Weeks after the treatment discontinuation will be considered as missing.

Descriptive statistics for the overall treatment effect based on the MMRM model will include the least-squares (LS) mean difference between each MD-7246 dose and placebo, LS mean change from baseline for each treatment, their corresponding 95% CIs, and the p-values associated with the comparisons to placebo. In addition, LS Means for the weekly treatment differences between each MD-7246 group and placebo will also be provided with the same set of statistics derived from the same MMRM model defined above and will be plotted based on the MMRM estimates.

Additionally, the non-model based descriptive statistics for the corresponding change from baseline (overall or weekly) will be provided by treatment group in the same tables for the MMRM estimates. Among them, the overall change from baseline is defined as the average of all non-missing post baseline daily scores during the Treatment Period minus the baseline abdominal pain score. The cumulative distribution for the overall change from baseline during the entire treatment period will be summarized and plotted by treatment group.

6/12 Week Abdominal Pain 30% Responder

For the 6/12 Week Abdominal Pain 30% Responder endpoint, the proportion of responders in each MD-7246 treatment group will be summarized and compared to the proportion of responders in the placebo group using a CMH test. The number and percentage of responders,

the differences in responder rates between each MD-7246 dose group and the placebo group, the odds ratio relative to placebo, the corresponding 95% CIs, and the associated p-values will be presented.

A bar plot for the percentage of 6/12 week 30% responders will be provided by treatment group based on the summary statistics. A forest plot for the odds ratios between active treatment arms and placebo will be provided with corresponding 95% confidence intervals.

Regarding the weekly 30% responders, the number and percentage of responders will be summarized for each week within Treatment Period, based on the mITT population. A bar plot by treatment group and by week (for Week 1 and Week 12 only) will be provided.

For any week, a patient with fewer than 4 daily scores available will be considered non-responder for that week.

7.8.9.3. Sensitivity Analyses of the Key Efficacy Endpoints

No sensitivity analysis analyses for the two key efficacy endpoints will be performed.

7.8.9.4. Additional Efficacy Endpoints and Analyses

The following endpoints are defined similarly to the Change from Baseline in Abdominal pain, for its overall and weekly. They will be analyzed using similar MMRM model and the non-model based summary statistics for the Change from Baseline in Abdominal Pain (through the Treatment Period and at each week):

- Change from Baseline in Abdominal Discomfort through the Treatment Period and at each week
- Change from Baseline in Abdominal Bloating through the Treatment Period and at each week
- Change from Baseline in Abdominal Cramping through the Treatment Period and at each week
- Treatment Satisfaction through Treatment period and at each week will be summarized and analyzed using MMRM model (without baseline in the model). Treatment Satisfaction is scored weekly on a 5-point scale during the Treatment Period only and no change from baseline is defined for this assessment.

The following 6/12 week responder endpoints are defined in the same manner as the 6/12 Week Abdominal Pain 30% Responder based on their relevant weekly responders. For each week in the Treatment Period, a weekly responder is a patient who satisfies the responder criteria in the change from baseline for that week. If a patient did not enter information into the eDiary on at least 4 days for a particular week during the Treatment Period, the patient will not be considered a weekly responder for that week. A patient who did not have any relevant post baseline assessment will be considered a weekly non-responder for all post baseline weeks, subsequently, will not be a 6/12 week responder. For all weeks after the treatment discontinuation, the patient will be considered as non-responder.

These responder endpoints will be summarized by treatment and the comparison between the active treatment group and placebo will be analyzed using a CMH test. The relevant number and percentage of weekly responders will be summarized by treatment at each week.

- 6/12 Week Abdominal Pain 40% Responder

  A 6/12 Week Abdominal Pain 40% Responder is a patient who meets the Weekly Abdominal Pain 40% Responder criteria for at least 6 out of the 12 weeks of the Treatment Period.

- 6/12 Week Abdominal Pain 50% Responder

  A 6/12 Week Abdominal Pain 50% Responder is a patient who meets the Weekly Abdominal Pain 50% Responder criteria for at least 6 out of the 12 weeks of the Treatment Period.

- 9/12 Week Abdominal Pain 30% Sustained Responder

  A 9/12 Week Abdominal Pain Sustained Responder is a patient who meets the Weekly Abdominal Pain 30% Responder criteria for at least 9 out of the 12 weeks of the Treatment Period.

- 9/12 Week Abdominal Pain 50% Sustained Responder

  A 9/12 Week Abdominal Pain Sustained Responder is a patient who meets the Weekly Abdominal Pain 50% Responder criteria for at least 9 out of the 12 weeks of the Treatment Period.

- 6/12 Week Abdominal Discomfort 30% Responder

  A 6/12 Week Abdominal Discomfort 30% Responder is a patient who meets the Weekly Abdominal Discomfort 30% Responder criteria for at least 6 out of the 12 weeks of the Treatment Period.

- 6/12 Week Abdominal Bloating 30% Responder

  A 6/12 Week Abdominal Bloating 30% Responder is a patient who meets the Weekly Abdominal Bloating 30% Responder criteria for at least 6 out of the 12 weeks of the Treatment Period.

- 6/12 Week Abdominal Cramping 30% Responder

  A 6/12 Week Abdominal Cramping 30% Responder is a patient who meets the Weekly Abdominal Cramping 30% Responder criteria for at least 6 out of the 12 weeks of the Treatment Period.

- 6/12 Week Degree of Relief Responder

  A 6/12 Week Degree of Relief Responder is a patient who scores ≤ 2 for at least 6 out of the 12 weeks of the Treatment Period.

- 6/12 Week Adequate Relief of IBS Pain Responder

A 6/12 Week Adequate Relief of IBS Pain Responder is a patient who reports adequate relief of IBS pain ("yes") for at least 6 out of the 12 weeks of the Treatment Period.

The following responder endpoints are defined based on whether a patient satisfies certain criteria for the change from baseline in daily abdominal pain at its worst on at least 50% of protocol defined treatment days (84 days). If a patient did not enter information into the eDiary on at least 42 days during Treatment Period, the patient will not be considered a responder for any of the following responder endpoints. Numbers and percentages of patients satisfying these responder definitions will be summarized by treatment and the comparison between each active treatment group and placebo will be analyzed using a CMH test.

- 50% of Days Daily Abdominal Pain 30% Responder

  A 50% of days Daily Abdominal Pain 30% Responder is defined as a patient who achieves at least 30% decrease from baseline in daily abdominal pain score at its worst for at least 50% of days in the 12-week Treatment Period, which is at least 42 days. These 50% days with at least 30% decrease don't have to be consecutive during treatment period.

- 50% of Days Daily Abdominal Pain 40% Responder

  A 50% of days Daily Abdominal Pain 40% Responder is defined as a patient who achieves at least 40% decrease from baseline in daily abdominal pain score at its worst for at least 50% of days in the 12-week Treatment Period, which is at least 42 days. These 50% days with at least 40% decrease don't have to be consecutive during the Treatment Period.

- 50% of Days Daily Abdominal Pain 50% Responder

  A 50% of days Daily Abdominal Pain 50% Responder is defined as a patient who achieves at least 50% decrease from baseline in daily abdominal pain score at its worst for at least 50% of days in the 12-week Treatment Period, which is at least 42 days. These 50% days with at least 50% decrease don't have to be consecutive during the Treatment Period.

Additionally, the cumulative distribution of patients satisfying 50% of days daily abdominal pain response based on the percent change from baseline will be plotted and tabulated by treatment group and by percent change from baseline in the daily abdominal pain at its worst.

Abdominal pain is measured daily using an 11-point NRS. Abdominal pain-free days will be those days where the patient reports a score of 0 for abdominal pain at its worst. The post baseline number of abdominal pain-free days per week will be calculated as the number of Abdominal Pain-free Days during the Treatment Period (from the first to the last dose), divided by the total number of days within Treatment Period times 7. The baseline percent number of abdominal pain-free days per week will be calculated as the number of Abdominal Pain-free Days during the Pretreatment Period (per defined Pretreatment Period), divided by the total number of days within the Pretreatment Period times 7. The descriptive statistics (n, mean, SD, median, minimum and maximum) for the baseline and post-baseline abdominal pain-free days

per week and the count of abdominal pain-free days during the Treatment Period will be tabulated by treatment group.

Assuming the count for the abdominal pain free days follows a Poisson distribution, the post-baseline abdominal pain-free days will be analyzed using a Poisson regression with treatment and baseline number of abdominal-pain free days as the covariates along with an offset variable of treatment duration (in weeks). In the case of overdispersion, the negative binomial model instead of Poisson regression will be implemented. Model estimates for weekly abdominal pain free days will be presented by treatment along with corresponding 95% confidence intervals. In the same table, the difference between the weekly rates for MD-7246 treatment arms and placebo will be represented with corresponding 95% confidence intervals and p-values associated with the comparisons to placebo.

7.8.10. Exploratory Endpoints and Analyses

The following endpoints will be summarized and analyzed via a similar MMRM for the Change from Baseline in Abdominal Pain through the Treatment Period and at each week:

- Change from Baseline in BM Frequency Rate through the Treatment Period and at each week

  For each period of interest (Pretreatment Period, Treatment Period, or a particular week), the BM frequency rate is a weekly rate, which is calculated as the total number of BM within the period divided by the total number of days during the period multiplied by 7. The change from baseline in BM frequency rate is the postbaseline frequency rate minus the baseline frequency rate. For weeks after the treatment discontinuation, this rate will be missing.

- Change from Baseline in BSFS (Stool Consistency) through the Treatment Period and at each week.

  For each period of interest (Pretreatment Period, Treatment Period, or a particular week), the BSFS is the average of all available BSFS scores the period. The change from baseline in BSFS is the postbaseline BSFS score minus the baseline score. For weeks after the treatment discontinuation, this score will be missing.

- Change from Baseline in the Percent of Days Experiencing an Episode of Diarrhea through the Treatment Period and at each week

  An episode of diarrhea is defined as a sequence of 2 or more loose/watery BMs (BSFS 6 or 7) that are never separated by >1 non-loose/watery stool or by a day without a BM. For each period of interest (Treatment Period, or a particular week), the percent of days experiencing an episode of diarrhea will be calculated as 100 times the number of days with a diarrhea episode divided by the total number of days with study treatment in the period. For baseline, the number of days with a diarrhea episode will be determined for the last 14 days of the Pretreatment Period. The baseline percent of days experiencing an episode of diarrhea will be calculated as 100 times the number of days experiencing an episode during the baseline period divided by the length in days of the baseline period (typically 14 days). The change from

baseline will be calculated for each week as the weekly percent minus the baseline percent. For weeks after the treatment discontinuation, this percent will be missing.

- Change from Baseline in the Percent of Days Experiencing No Episodes of Diarrhea through the Treatment Period and at each week

  For each period of interest, the Percent of Days experiencing no episode of diarrhea will be 100% minus the percent of days experiencing episode of diarrhea.

- Change from Baseline in BM-related Symptom Severity through the Treatment Period

  Patient global impression of severity (PGI-S) of BM-related symptom severity on a 5-point scale will be reported weekly by eDiary entry. Weekly PGI-S will be summarized weekly and analyzed via MMRM.

- BM-related Symptom Change

  Patient global impression of change (PGI-C) of BM-related symptoms is measured weekly using a 7-point ordinal scale comparing the severity for the past 7 days to the severity before starting the study. The postbaseline weekly PGI-C score will be analyzed via MMRM (without baseline).

Urgent BM Frequency Rate and Number of Days with Use of Loperamide for Diarrhea per week will be defined similarly as Number of Abdominal Pain-free days per week and will be calculated as the total number of events within the entire Treatment Period (or Pretreatment Period for baseline) divided by the total number of days within the Treatment Period, then multiplied by 7. Similar to Number of Abdominal Pain-free Days per week, these endpoints will be summarized by treatment group and the relevant count data during the entire treatment period will be analyzed by Poisson regression for the comparison between any active treatment group and placebo. If the data is skewed to 0 (overdispersed), the Negative binomial model will be used.

7.8.11. Safety Analysis

All safety analyses will be performed using the Safety Population.

7.8.11.1. Adverse Events

Adverse event verbatim terms will be coded in the EDC system against Medical Dictionary for Regulatory Activities (MedDRA) version 22.0.

During the TE Period, an AE that has a start date during the TE Period (from the date of the first dose to 1 day after the last dose of double-blind study drug) will be considered a TEAE. For the TE Period, the number and percentage of patients reporting TEAEs in each treatment group will be tabulated:

- By system organ class (SOC) and preferred term (PT)
- By SOC, PT, and severity
- By SOC, PT, and relationship to study drug

If a patient has more than one TEAE coded to the same PT or SOC, the patient will be counted only once for that PT or SOC by identifying those TEAEs with the highest severity and the closest relationship to study drug.

The incidence of the following TEAEs will be summarized by PT:

- Common (≥2% of patients in any treatment group) TEAEs
- Treatment-emergent SAEs
- TEAEs leading to premature discontinuation of study drug

In addition, the incidence of SAEs, fatal SAEs (ie, events with an outcome of death), if any, will be summarized separately by treatment group and PT within the period from the first dose to 30 days after the last dose of study drug.

AE Listings will be presented for all patients with AEs, screened patients with SAEs, patients with AEs leading to discontinuation, and patients who died (if any), respectively.

7.8.11.2. Clinical Laboratory Parameters

Descriptive statistics for clinical laboratory values (in standard units) and changes from the baseline values at each assessment time point will be presented by treatment group, for each clinical laboratory parameter.

The number and percentage of patients with potentially clinically significant (PCS) post-baseline clinical laboratory values will be tabulated by treatment group for the TE period. The criteria for PCS laboratory values are described in Table 2. The percentages will be calculated relative to the number of patients with available non-PCS baseline values and at least 1 assessment in the corresponding post-baseline TE period. The numerator will be the total number of patients with available non-PCS baseline values and at least 1 PCS value in the corresponding post-baseline period. A supportive listing of patients with PCS post-baseline values will be provided, including the PID number, study center, and baseline and post-baseline values. A listing of all AEs for patients with PCS laboratory values will also be provided.

Table 2: Criteria for Potentially Clinically Significant Laboratory Results

Parameter	*SI Unit*	*Lower Limit*	*Higher Limit*
CHEMISTRY			
Albumin	g/L	< 0.9 × LLN	> 1.1 × ULN
Alanine aminotransferase	U/L	—	≥ 3 × ULN
Alkaline phosphatase	U/L	—	≥ 3 × ULN
Aspartate aminotransferase	U/L	—	≥ 3 × ULN
Bicarbonate	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Bilirubin, total	µmol/L	—	> 1.5 × ULN
Calcium	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Chloride	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Cholesterol, total	mmol/L	—	> 1.6 × ULN

Parameter	*SI Unit*	*Lower Limit*	*Higher Limit*
Creatinine	µmol/L	—	> 1.3 × ULN
Glucose	mmol/L	< 0.8 × LLN	> 1.4 × ULN
Magnesium	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Phosphate	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Potassium	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Protein, total	g/L	< 0.9 × LLN	> 1.1 × ULN
Sodium	mmol/L	< 0.9 × LLN	> 1.1 × ULN
Urea nitrogen	mmol/L	—	> 1.2 × ULN
Uric acid	µmol/L	< 0.9 × LLN	> 1.1 × ULN
HEMATOLOGY			
Basophils, absolute cell count	10^9/L	—	> 3 × ULN
Eosinophils, absolute cell count	10^9/L	—	> 3 × ULN
Hematocrit	Ratio	< 0.9 × LLN	> 1.1 × ULN
Hemoglobin	g/L	< 0.9 × LLN	> 1.1 × ULN
Lymphocytes, absolute cell count	10^9/L	< 0.8 × LLN	> 1.5 × ULN
Mean corpuscular hemoglobin	pg	—	> 3 × ULN
Mean corpuscular hemoglobin concentration	g/L	—	> 3 × ULN
Mean corpuscular volume	fL	< 0.9 × LLN	> 1.1 × ULN
Monocytes, absolute cell count	10^9/L	—	> 3 × ULN
Neutrophils, absolute cell count	10^9/L	< 0.8 × LLN	> 1.5 × ULN
Platelet count	10^9/L	< 0.5 × LLN	> 1.5 × ULN
Red blood cell count	10^{12}/L	< 0.9 × LLN	> 1.1 × ULN
White blood cell count	10^9/L	< 0.7 × LLN	> 1.5 × ULN

LLN=lower limit of normal value provided by the laboratory; SI=*Le Système International d'Unités* (International System of Units); ULN=upper limit of normal value provided by the laboratory.

7.8.11.3. Vital Signs

Descriptive statistics for vital signs (ie, pulse rate, systolic and diastolic BP, respiratory rate, temperature, and body weight) and changes from baseline at each visit and at the end of the period/study will be presented by treatment group.

The number and percentage of patients with PCS post-baseline vital signs will be tabulated by treatment group for the Treatment Emergent Period. A vital sign value will be considered PCS if it meets both the observed value criterion and the change from baseline value criterion. The criteria for PCS vital sign values is detailed in Table 3. The percentages will be calculated relative to the number of patients with available non-PCS baseline values and at least 1 assessment in the corresponding postbaseline period. The numerator will be the total number of patients with available baseline values and at least 1 PCS value in the corresponding post-baseline period. A supportive listing of patients with PCS post-baseline values will be provided, including the PID number, study center, and baseline and post-baseline values. A listing of all AEs for patients with PCS vital sign values will also be provided.

Table 3: Criteria for Potentially Clinically Significant Vital Signs

Parameter	***Flag***	***Criteria[a]***	
		Observed Value	***Change from Baseline***
Sitting systolic blood pressure, mmHg	High	≥ 180	Increase of ≥ 20
	Low	≤ 90	Decrease of ≥ 20
Sitting diastolic blood pressure, mmHg	High	≥ 105	Increase of ≥ 15
	Low	≤ 50	Decrease of ≥ 15
Sitting pulse rate, bpm	High	≥ 120	Increase of ≥ 15
	Low	≤ 50	Decrease of ≥ 15
Weight, kg	High	—	Increase of ≥ 7%
	Low	—	Decrease of ≥ 7%

a A post-baseline value is considered potentially clinically significant if it meets both the observed-value and the change-from-baseline criteria.
bpm=beats per minute.

7.8.11.4. Physical Examination

Any physical examination abnormality that the investigator considers to be potentially clinically significant and changed from the baseline will be reported as an AE. No separate analysis for Physical Examinations is planned.

7.8.12. Health Outcomes Analysis

Pain Catastrophizing Scale

The Pain Catastrophizing Scale is a 13-item instrument that asks patients to reflect on their thoughts and feelings of past painful experiences. The patient responds to each statement using a 5-point scale (0=None at all to 4=All the time). The total Pain Catastrophizing Scale score plus the 3 subscale scores (Rumination, Magnification, and Helplessness) will be derived and summarized by treatment for the Pretreatment Visit based on mITT population.

IBS-SSS

The IBS-SSS parameters consist of 5 individual items and the IBS-SSS total score. The 5 individual items (severity of abdominal pain, frequency of abdominal pain, severity of abdominal distension, dissatisfaction with bowel habits, and interference of IBS with life in general) are based on a 0-100 scale (10 times the original scale) and the IBS-SSS total score parameter is the sum of the 5 IBS-SSS individual item scores (0-500 scale).

For each post-baseline visit, change from baseline in the IBS-SSS total score and individual items will be summarized analyzed using an MMRM model similar to that for the change from baseline in abdominal pain.

SF-12v2 and EQ-5D-3L

SF-12v2 and EQ-5D-3L analyses will not be included in this statistical analysis plan, but will be discussed in a separate analysis plan.

8. REFERENCES

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APPENDIX 1. CHANGES FROM ANALYSES PLANNED IN THE PROTOCOL

Type	Protocol	SAP	Justifications
Language Clarification	Change from baseline at Each Week in various efficacy assessments	Change from baseline through the Treatment Period and at Each Week	To differentiate • Change through the entire Treatment Period. • Individual weekly changes
	Treatment Period was used for 2 periods: • From the day of the first dose to 1 day after the last dose • From the day of the first dose to the day of the last dose	Clearly distinguish different periods: • From the day of the first dose to 1 day after the last dose is defined as Treatment Emergent Period. • From the day of the first dose to the day of the last dose is defined as Treatment Period.	Clarification
Operational Adjustment	• An optional interim analysis of unblinded safety and efficacy data may be performed to assess futility. • Sensitivity analyses planned for the change from baseline in abdominal pain score • Analyses for Health outcome parameters SF-12v2 and EQ-5D-3L were included.	• No interim analysis performed • No sensitivity analysis performed • Analyses for Health outcome parameters SF-12v2 and EQ-5D-3L will be in a separate analysis plan.	• Faster enrollment won't leave time for an interim analysis. • In a hypothesis generating phase 2 study, the sensitivity analysis is unnecessary. • HEOR parameters will be detailed out in relevant analysis plan

Type	Protocol	SAP	Justifications
Methodologic Changes	• MMRM analysis for the abdominal pain change from baseline included geographic region. • Estimation, terminology and analysis for o Percent of Abdominal Pain Free Days o Urgent BM Frequency Rate o Percent of Days with Use of Loperamide for Diarrhea • The quantities were estimated for each week, and analyzed by MMRM • Analysis for IBS-SSS total score and sub-scores was based on ANCOVA model.	• MMRMs did not adjust for geographic region. • New Terms o Abdominal Pain Free Days per week o Urgent BM Frequency Rate o Number of Days with Use of Loperamide for Diarrhea • Quantities are estimated for the Treatment period and from Poisson regression model using the relevant count data • IBS-SSS total score and sub-scores was based on MMRM model	• Sample size limited number of covariates in the MMRM model. • Relatively rare events for patients in IBS-D lead to different statistical distribution for the count data • Multiple visits in IBS-SSS support repeated measure analysis
Addition	The following endpoints and relevant analyses were not included: • 6/12 week abdominal pain at its worst 40% responder • 9/12 week abdominal pain at its worst 30, 50% sustained responders • 50% days 30%, 40%, 50% responders	The following endpoints and relevant analyses are included: • 6/12 week abdominal pain at its worst 40% responder • 9/12 week abdominal pain at its worst 30, 50% sustained responders • 50% days 30%, 40%, 50% responders	Added for exploratory purposes.

APPENDIX 2. VISIT TIME WINDOWS FOR SAFETY ANALYSIS

Appendix Table 2-1 below presents the visits assigned for the safety analysis corresponding to the range of trial days (window) during which an actual visit may have occurred.

Appendix Table 2-1: Visit Time Windows for Safety Analysis

Derived Visit	***Scheduled Test / Visit Day[a]***	***Window***
Baseline	Day 1	Days ≤ 1
Week 4 (Day 29) Visit	Day 29 (+/- 3 days)	Days [2, 43]
Week 8 (Day 57) Visit	Day 57 (+/- 3 days)	Days [44, 71]
Week 12 (Day 85) Visit	Day 85 (+ 3 days)	Days ≥ 72
End of Treatment	Day of Last Study Treatment based on End of Treatment CRF form	
End of Study	Study Day for the End of Study Follow-Up Phone Call	

a Relative to the date of first dose (Day 1).
b "End of Treatment/End of Study" will be presented in analysis tables for safety parameters, including ECG, clinical laboratory, and vital signs.

Test/Visit Day will be calculated as follows: test/visit date – date of first dose + 1.

If there are multiple values available for the same Visit, the data with the latest date for that visit will be utilized for summary.

APPENDIX 3. VISIT TIME WINDOWS FOR EFFICACY ANALYSIS

Appendix Table 3-1 presents the analysis weeks assigned for the efficacy analysis of the patient diary data related to BM characteristics. These analysis weeks will be used in the calculations for all week-based parameters (eg, abdominal pain, BSFS weekly scores).

Appendix Table 3-1: Analysis Time Windows for Efficacy Analysis

Period	*Analysis Week*	*Begins*[a]	*Ends*[a]
Pretreatment (Baseline[b])	Week –2	Day –14	Day –8
	Week –1	Day –7	Day 1 (time of first dose)
Treatment	Week 1	Day 1 (time of first dose)	Day 7
	Week 2	Day 8	Day 14
	Week 3	Day 15	Day 21
	Week 4	Day 22	Day 28
	Week 5	Day 29	Day 35
	Week 6	Day 36	Day 42
	Week 7	Day 43	Day 49
	Week 8	Day 50	Day 56
	Week 9	Day 57	Day 63
	Week 10	Day 64	Day 70
	Week 11	Day 71	Day 77
	Week 12	Day 78	Day of last study treatment dose[c] (usually Day 85)

Note: There is no Day 0 or Week 0. For eDiary assessments in which a patient is asked to report if an event occurred "yesterday" or "today" (e.g., a bowel movement), these windows pertain to when the event occurred, not to when the event is reported. For example, if a patient reports in eDiary on Day 78 a bowel movement occurring "yesterday," that bowel movement (and subsequent stool consistency) would be included in Analysis Week 11, not Analysis Week 12.

a Relative to the date of first dose; Day 1 = the day of first dose. For the calculation of rates in which the duration of a week or the overall period is calculated to the nearest hour, a day begins and ends at midnight.

b Baseline values for efficacy parameters will be derived from the daily eDiary and eCRF data collected in the Pretreatment Period, specifically the period from 14 days before Randomization Visit up to the time of first dose.

c For patients who fail to provide the date of last dose n the End of Treatment CRF form, the later date of the last known dose (drug dispense eCRF) or the last eDiary date will be used to impute the last dose date.

For the Treatment Period, diary day is calculated as diary date – date of first dose + 1. For the Pretreatment Period, diary day is calculated as diary date - date of first dose. However, the day of first dose is always trial Day 1.

If a patient withdraws during the Treatment Period, the patient's Treatment Period shall end on the day of the last dose. The affected Treatment Period shall be shortened to the end of the withdrawn patient's Treatment Period, and all subsequent weeks will be missing for that patient.

Appendix Table 3-2 presents the analysis weeks assigned for the efficacy analysis of the patient diary data related to Weekly Questions (eg, Weekly Patient Assessment of Degree of Relief of IBS Symptoms, Weekly Patient Assessment of Adequate Relief of IBS Pain, and Weekly Patient Assessment of Treatment Satisfaction).

Appendix Table 3-2: Analysis Time Windows for Efficacy Analysis – Weekly Assessments

Period	*Analysis Week*	*Begins*[a]	*Ends*[a]
Pretreatment (Baseline[b])	Week –2	Day –11	Day –5
	Week –1	Day –4	Day 1 (time of first dose)
Treatment	Week 1	Day 7	Day 10
	Week 2	Day 11	Day 17
	Week 3	Day 18	Day 24
	Week 4	Day 25	Day 31
	Week 5	Day 32	Day 38
	Week 6	Day 39	Day 45
	Week 7	Day 46	Day 52
	Week 8	Day 53	Day 59
	Week 9	Day 60	Day 65
	Week 10	Day 66	Day 72
	Week 11	Day 73	Day 79
	Week 12	Day 80	3 Days after last study treatment dose (usually Day 84)*

a Relative to the date of first dose; Day 1 = the day of first dose. For the calculation of rates in which the duration of a week or the overall period is calculated to the nearest hour, a day begins and ends at midnight.

b Baseline values for efficacy parameters will be derived from the weekly eDiary and eCRF data collected in the Pretreatment Period.

In general, weekly questions will be assigned only to the analysis week for which the question covers at least 4 days of that week.

If a weekly question covers days from different study phases, the answers will be discarded. For example, a weekly question answered on study day 2 would cover part of 1 day from the Treatment Period and 6 days from the Pretreatment Period, so these data would be discarded.

If patients answer the weekly questions multiple times for a week, for each question, the average of the respective answers will be assigned as the weekly value.

For time periods covering more than a week, the value will be calculated as the average of the weekly values. Thus, for Pretreatment, the value will be calculated as the average of week -2 and week -1.

APPENDIX 4. BRISTOL STOOL FORM SCALE

Type 1 - Separate hard lumps like nuts (difficult to pass)

Type 2 - Like a sausage but lumpy

Type 3 - Like a sausage but with cracks on the surface

Type 4 - Like a sausage or snake, smooth and soft

Type 5 - Soft pieces with clear-cut edges (easy to pass)

Type 6 - Fluffy pieces with ragged edges, a mushy stool

Type 7 - Watery, no solid pieces (entirely liquid)