Clinical Trial Protocol: MCP-103-312

Final Version, 26 March 2018

Study Title:	A Phase 3b, Randomized, Double-blind, Placebo-controlled, Parallel-group Trial of Linaclotide 290 µg Administered Orally for 12 Weeks Followed by a 4-week Randomized Withdrawal Period in Patients with Irritable Bowel Syndrome with Constipation
Study Number:	MCP-103-312
Study Phase:	3b
Product Name:	Linaclotide
Indication:	Irritable Bowel Syndrome with Constipation
Investigators:	Multicenter
Sponsor:	Ironwood Pharmaceuticals, Inc. 301 Binney Street Cambridge, MA 02142
Sponsor Contact:	[redacted]
Medical Monitor:	[redacted]

	Date
Original Protocol:	26 March 2018

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SYNOPSIS

Trial Number:

MCP-103-312

Trial Title:

A Phase 3b, Randomized, Double-blind, Placebo-controlled, Parallel-group Trial of Linaclotide 290 µg Administered Orally for 12 Weeks Followed by a 4-week Randomized Withdrawal Period in Patients with Irritable Bowel Syndrome with Constipation

Trial Centers:

Approximately 80 in the United States

Development Phase: 3b

Objective(s):

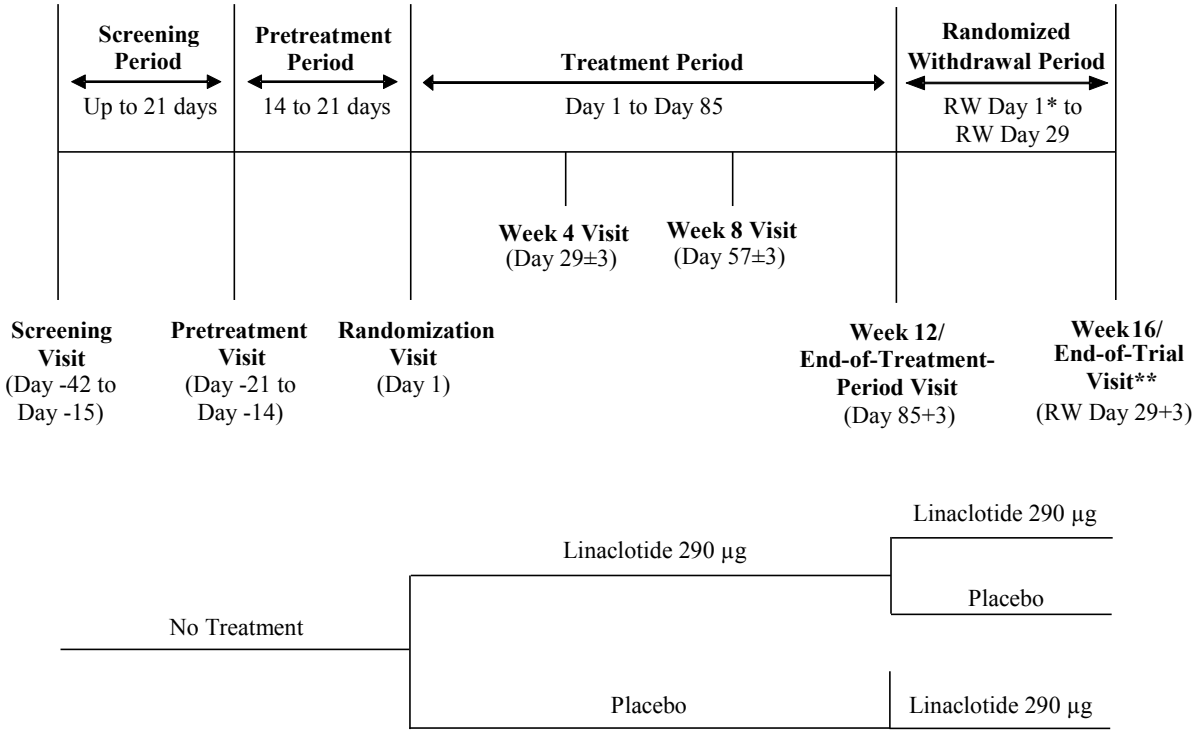
To evaluate the efficacy on abdominal symptoms (abdominal bloating, abdominal discomfort, and abdominal pain) and safety of linaclotide 290 µg administered orally to patients with irritable bowel syndrome with constipation (IBS-C).

Methodology:

This is a multicenter, randomized, double-blind, placebo-controlled, parallel-group trial, consisting of 4 distinct periods, as illustrated in the figure below. The trial will enroll patients who have IBS-C diagnosed using Rome III criteria. Eligible patients will be randomized to 1 of 2 treatments: linaclotide 290 µg or placebo (1:1) once daily. Patients who complete the 12-week Treatment Period will enter the 4-week Randomized Withdrawal (RW) Period and be allocated to study drug in a double-blind manner, as follows:

- Patients randomized to linaclotide 290 µg during the Treatment Period will be rerandomized to linaclotide 290 µg or placebo (1:1)
- Patients randomized to placebo during the Treatment Period will be allocated to linaclotide 290 µg

Overview of Trial Design



Note: there is no Day 0.
RW=Randomized Withdrawal

* RW Day 1 is the day after the Week 12/End-of-Treatment-Period Visit.

** This visit represents the end of the trial.

Trial Periods

1. Screening Period: The Screening Period starts with the signing of the informed consent form (ICF; Appendix 1) and may last for up to 21 days. During this period, patient eligibility for entry into the Pretreatment Period will be determined. 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 (see Appendix 2), the Screening Visit and Pretreatment Visit may be combined into one visit.
2. 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 Bowel Movement (BM)-related Symptom-severity Assessments on an event-driven basis (meaning these are assessments made at the time the event occurs)

- Daily Abdominal Symptom-severity Assessments in an evening report
- Weekly Patient Assessment of Constipation Severity
- Weekly Patient Assessment of IBS Symptom Severity
- Weekly Patient Assessment of Adequate Relief
- Use of Bisacodyl or Other Laxatives, Suppositories, or Enemas on an event-driven basis

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

3. Treatment Period: The Treatment Period begins with treatment assignment and lasts for 12 weeks. Patients will be randomly assigned to 1 of 2 treatments: linaclotide 290 µg or placebo. Patients will take their initial dose of study drug at the trial center during the Randomization Visit, after fasting for 2 hours. On all other days, study drug will be taken once daily in the morning at least 30 minutes before breakfast. Patients will continue to use the handheld eDiary to provide their Daily BM-related and Abdominal Symptom-severity Assessments, weekly assessments (as described above and including Weekly Patient Assessment of Treatment Satisfaction), and Use of Bisacodyl or Other Laxatives, Suppositories, or Enemas. Other patient-reported outcomes assessments (IBS Quality of Life Questionnaire [IBS-QOL] and CDC Healthy Days Core Module [CDC HRQOL-4]) will be administered at trial visits throughout the Treatment Period. Patients will complete a Week 4 Visit, Week 8 Visit, and Week 12/End-of-Treatment-Period (ETP) Visit during the Treatment Period (see Schedule of Evaluations).
4. Randomized Withdrawal (RW) Period: The RW Period is defined as the 4 weeks immediately following the Treatment Period. Patients who complete the 12-week Treatment Period and return for the Week 12/ETP Visit will enter the 4-week RW Period and be allocated to study drug in a double-blind manner, as follows:
   - Patients randomized to linaclotide 290 µg during the Treatment Period will be rerandomized to linaclotide 290 µg or placebo (1:1)
   - Patients randomized to placebo during the Treatment Period will be allocated to linaclotide 290 µg

   Patients will take their first dose of study drug in the RW Period the day after the Week 12/ETP Visit (this will be considered RW Day 1). As during the Treatment Period, study drug will be taken once daily in the morning at least 30 minutes before breakfast. Patients will continue to use the handheld eDiary to provide their daily assessments, weekly assessments, and laxative use. Other patient-reported outcomes will be administered at the trial visit during the RW Period. At the end of the RW Period, patients will complete the Week 16/End-of-Trial (EOT) Visit.

Trial Procedures

During the Pretreatment, Treatment, and RW 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.

Daily Assessments: The following information will be entered into the eDiary each day:

- Diary for IBS Symptoms for Constipation-Predominant (DIBSS-C)
  - Daily BM-related Symptom-severity Assessments (entered by the patient for each BM on an event-driven basis):
    - BM day and time
    - Association with a sense of a complete evacuation (yes/no)
    - Stool consistency using the Bristol Stool Form Scale (BSFS; 1=Separate hard lumps like nuts [difficult to pass] to 7=Watery, no solid pieces [entirely liquid])
    - Straining on a 5-point ordinal scale (1=Not at all to 5=An extreme amount [extremely severe straining])
    - Stool consistency using a 5-point ordinal scale (1=Very hard to 5=Very loose and watery)
  - Daily Abdominal Symptom-severity Assessments (entered by the patient in a daily evening report):
    - Rating of abdominal pain at its worst during the previous 24 hours on an 11-point numerical rating scale (NRS; 0=No abdominal pain to 10=Worst possible abdominal pain)
    - Rating of abdominal discomfort at its worst during the previous 24 hours on an 11-point NRS (0=No abdominal discomfort to 10=Worst possible abdominal discomfort)
    - Rating of abdominal bloating at its worst during the previous 24 hours on an 11-point NRS (0=No abdominal bloating to 10=Worst possible abdominal bloating)
- Use of Bisacodyl or Other Laxatives, Suppositories, or Enemas
  - The day and time will be entered by the patient for each use of protocol-permitted bisacodyl (tablets or suppositories) or other laxatives, suppositories, or enemas on an event-driven basis

Weekly Assessments: The following information will be entered by the patient into the eDiary once per week at the same time as the daily evening report:

- Weekly Patient Assessment of Constipation Severity (5-point ordinal scale; 1=None to 5=Very severe)
- Weekly Patient Assessment of IBS Symptom Severity (5-point ordinal scale; 1=None to 5=Very severe)
- Weekly Patient Assessment of Adequate Relief (yes/no)
- Weekly Patient Assessment of Treatment Satisfaction (5-point ordinal scale; 1=Not at all satisfied to 5=Very satisfied; captured each week after the Randomization Visit)

Trial Visit Assessments: The following information will be captured in the eDiary at visits during the Treatment Period and RW Period:

- IBS Quality of Life Questionnaire (IBS-QOL, captured at the Randomization Visit before the first dose of study drug and all subsequent trial visits)
- CDC Healthy Days Core Module (CDC HRQOL-4, captured at the Randomization Visit before the first dose of study drug, the Week 12/ETP Visit, and the Week 16/EOT Visit)

Concomitant Medicine

- Rescue Medicine: During the Pretreatment, Treatment, and RW Periods, patients may use dispensed, protocol-permitted laxatives (bisacodyl, as tablets or suppositories) as Rescue Medicine when at least 72 hours have passed since their previous BM or when their symptoms become intolerable. In order to qualify for randomization into the Treatment Period, patients must not have used Rescue Medicine on the day before the Randomization Visit or on the day of the Randomization Visit up until the time of the clinic visit. Patients must agree to refrain from using Rescue Medicine from the time they arrive at the clinic for the Randomization Visit through the day after randomization.
- Prohibited Medicine: Medicines that are not permitted during the Pretreatment, Treatment, and RW Periods are listed in Appendix 2.

Number of Patients:

Approximately 600 IBS-C patients (300 patients per treatment group) will be randomized into the Treatment Period.

Diagnosis and Main Criteria for Inclusion:

Inclusion Criteria

To be eligible to participate in the trial, patients must meet the following criteria:

1. Patient has given consent by signing an ICF.
2. Patient is ambulatory (females must also be non-pregnant) and is aged 18 years or older at the Screening Visit. Lactating females must agree not to breastfeed.
3. Female patients of childbearing potential (ie, women who are not postmenopausal or who have not had a bilateral oophorectomy, hysterectomy, or tubal ligation) who are sexually active with a male partner must agree to use one of the following methods of birth control from the date they sign the ICF until 24 hours after their final dose of study drug:
   a. Hormonal contraception (ie, oral contraceptive, contraceptive implant, or injectable hormonal contraceptive)
   b. Double-barrier birth control (eg, condom plus intrauterine device, diaphragm plus spermicide)
   c. Maintenance of a monogamous relationship with a male partner who has been surgically sterilized by vasectomy
4. Females of childbearing potential must have a negative serum pregnancy test at the Screening Visit and a negative urine pregnancy test at the Randomization Visit prior to dosing.
5. Patient meets the colonoscopy requirements as defined by the American Gastroenterological Association guidelines and described in Appendix 3. (Note: Patients who receive narcotic anesthesia for a colonoscopy are eligible to enter the Pretreatment Period on the fifth day after the colonoscopy.)
6. Patient meets Rome III criteria for IBS (1): reports abdominal pain or discomfort at least 3 days/month during the 3 months before the diagnosis with the onset at least 6 months before the diagnosis, associated with two or more of the following features:
   a. Improved with defecation
   b. Onset associated with a change in frequency of stool
   c. Onset associated with a change in form (appearance) of stool

   Note: the diagnosis can be made at the Screening Visit or can be based on symptoms that the patient had before starting chronic treatment with linaclotide (LINZESS®), lubiprostone (AMITIZA®), or plecanatide (TRULANCE™).
7. During the time when the Rome III criteria for IBS were met, the patient had hard or lumpy stools (Bristol Stool Form Scale [BSFS] scores of 1 or 2) with at least 25% of BMs and had loose (mushy) or watery stools (BSFS scores of 6 or 7) with less than 25% of BMs in the absence of antidiarrheal or laxative use.

8. Patient reports <3 BMs (with each BM occurring in the absence of any laxative, suppository, or enema use during the preceding 24 hours) per week for at least 12 weeks, before the Screening Visit or before starting chronic treatment with linaclotide (LINZESS®), lubiprostone (AMITIZA®), polyethylene glycol 3350 (MiraLAX®), plecanatide (TRULANCE™), or any laxative.
9. Patient has an average score for abdominal pain at its worst of ≥3.0, as reported in the eDiary using an 11-point numerical rating scale (NRS) during the 14 days before the Randomization Visit and including the data entry into the eDiary made in clinic at the Randomization Visit prior to randomization.
10. Patient reports ≤6 complete spontaneous BMs (CSBMs) and ≤10 spontaneous BMs (SBMs) in the eDiary occurring over the 14 days before the Randomization Visit and including the data entry into the eDiary made in clinic at the Randomization Visit prior to randomization. (Note: A CSBM is an SBM that is associated with a sense of complete evacuation. An SBM is a BM that occurs in the absence of laxative, suppository, or enema use on the calendar day of the BM or the calendar day before the BM as evidenced by the patient-reported laxative use via the eDiary.)
11. Patient is compliant (as defined in the trial procedures section below) with eDiary completion by adequately responding to eDiary questions (ie, completing the daily evening report) on 10 or more of the 14 days before the Randomization Visit.
12. Patient is willing to discontinue any laxatives used before the Pretreatment Visit in favor of the protocol-defined Rescue Medicine (bisacodyl tablets or suppositories).
13. Patient is fluent in English or Spanish.
14. Patient agrees to refrain from making any new, major life-style changes that may affect IBS-C symptoms (eg, starting a new diet or changing his or her exercise pattern) from the time of signature of the ICF to the last trial visit.

Exclusion Criteria

Patients who meet any of the following criteria will not be eligible to participate in the trial:

1. Patient reports loose (mushy) or watery stools (BSFS score of 6 or 7) in the absence of any laxative, suppository, enema, or prohibited medicine (as described in Appendix 2) for >25% of BMs during the 12 weeks before the Screening Visit.
2. Patient has clinically significant concurrent illness or findings on a physical examination or clinical laboratory tests (clinical chemistry panel, complete blood count [CBC], urine drug screen) after signing the ICF but before receiving the first dose of study drug. (Note: The investigator will determine if a particular finding is clinically significant. In making this determination, the investigator will consider whether the particular finding could prevent the patient from performing any of the protocol-specified assessments, could

represent a condition that would exclude the patient from the trial, could represent a safety concern if the patient participates in the trial, or could confound the trial-specified assessments of safety or efficacy.)

3. Patient has been diagnosed with or has a family history of familial adenomatous polyposis, hereditary nonpolyposis colorectal cancer, or any other form of familial colorectal cancer.
4. Patient has a prior history of inflammatory bowel disease (IBD).
5. Patient currently has clinically significant symptoms such as lower GI bleeding (rectal bleeding or heme-positive stool), iron-deficiency anemia, weight loss, or systemic signs of infection or colitis.
6. Patient currently has active peptic ulcer disease (ie, not adequately treated or stable with therapy).
7. Patient has a history of diverticulitis or any chronic condition (eg, chronic pancreatitis, polycystic kidney disease, ovarian cysts, endometriosis) with ongoing symptoms that can be associated with abdominal pain or discomfort and could confound the assessments in this trial.
8. Patient has a central nervous system cause of constipation (eg, Parkinson's disease, spinal cord injury, and multiple sclerosis).
9. Patient has ever had any of the following diseases or conditions that can be associated with constipation: pseudo-obstruction, megacolon, megarectum, bowel obstruction, descending perineum syndrome, solitary rectal ulcer syndrome, systemic sclerosis.
10. Patient has ever had a fecal impaction that required hospitalization or emergency room treatment, or has a history of cathartic colon, laxative or enema abuse, ischemic colitis, or pelvic floor dysfunction (unless successful treatment has been documented by a normal balloon expulsion test).
11. Patient has a known or suspected structural abnormality of the gastrointestinal (GI) tract (eg, mechanical gastrointestinal obstruction) or a disease or condition that can affect GI motility.
12. Patient has a history of hypersensitivity to linaclotide or to any of the excipients contained in the study drug (active or placebo) as described in Section 7.1.
13. Patient has had surgery that meets any of the following criteria:
    a. Bariatric surgery for treatment of obesity, or surgery to remove a segment of the GI tract at any time before the Screening Visit
    b. Surgery of the abdomen, pelvis, or retroperitoneal structures during the 6 months before the Screening Visit
    c. An appendectomy or cholecystectomy during the 60 days before the Screening Visit
    d. Other major surgery during the 30 days before the Screening Visit

14. Patient has a history of cancer other than treated basal cell or squamous cell carcinoma of the skin. (Note: Patients with a history of cancer are allowed if the malignancy has been in a complete remission for at least 5 years before the Randomization Visit. A complete remission is defined as the disappearance of all signs of cancer in response to treatment.)
15. Patient has a history of diabetic neuropathy.
16. Patient has untreated hypothyroidism or treated hypothyroidism for which the dose of thyroid hormone has not been stable for at least 6 weeks at the time of the Screening Visit.
17. Patient has a recent history (during the 12 months before the Randomization Visit) of drug or alcohol abuse. (Note: Patients with a history of drug or alcohol abuse that was diagnosed greater than 12 months before the Randomization Visit may be enrolled if they have exhibited no actual abuse during the 12 months before the Randomization Visit.)
18. Patient reports a BSFS score of 6 (loose, mushy stools) for >1 SBM or a BSFS score of 7 (watery stools) with any SBM occurring over the 14 days before the Randomization Visit and including the data entry into the eDiary made in the clinic at the Randomization Visit prior to randomization.
19. Patient used Rescue Medicine (bisacodyl tablet or suppository) or any other laxative, suppository, or enema, on the day before or the day of the Randomization Visit prior to randomization.
20. Patient reported using a Prohibited Medicine (excluding laxatives, suppositories, and enemas) during the Pretreatment Period or is not willing or able to abide by the restrictions regarding use of Prohibited Medicines defined in Appendix 2. (Note: The use of fiber, bulk laxatives, or stool softeners [such as docusate] is acceptable provided the patient has been on a stable dose during the 30 days before the Screening Visit and plans to continue on a stable dose throughout the trial.)
21. Patient has been hospitalized for a psychiatric condition or has made a suicide attempt during the two years before the Randomization Visit.
22. Patient has taken commercially available linaclotide (LINZESS®) or plecanatide (TRULANCE™) or received an investigational drug during the 30 days before the Screening Visit.
23. Patient has an acute or chronic condition that, in the investigator's opinion, would limit the patient's ability to complete or participate in this clinical trial.
24. Patient has previously entered the Pretreatment Period of this trial.
25. Patient is directly or indirectly involved in the conduct and administration of this trial as an investigator, sub-investigator, study coordinator, other trial staff member, or employee of Ironwood Pharmaceuticals, Forest Laboratories, Actavis, or Allergan; or the patient is a first-degree family member, significant other, or relative residing with one of the above

persons involved directly or indirectly in the trial; or the patient is enrolled in this trial at another clinical trial site.

Test Product, Dosage, and Mode of Administration:

Linaclotide 290 µg oral capsule administered in a double-blind manner once daily in the morning at least 30 minutes before breakfast.

Reference Therapy, Dosage, and Mode of Administration:

Matching placebo oral capsule administered in a double-blind manner once daily in the morning at least 30 minutes before breakfast.

Duration of Treatment:

The test product and reference therapy will be administered for 16 weeks. Total patient participation is expected to last for up to 162 days.

Criteria for Evaluation:

Primary Efficacy Assessments

The following efficacy assessments entered in the eDiary are used to determine the primary efficacy endpoint: daily patient assessments of abdominal bloating, abdominal discomfort, and abdominal pain at their worst over the last 24 hours (each on an 11-point NRS).

Additional Efficacy Assessments

In addition to the primary efficacy assessments, additional efficacy assessments are based on daily assessments of SBMs and CSBMs (including number and day/time of BMs, laxative use, and completeness of evacuation), stool consistency as measured on both the 7-point BSFS and 5-point ordinal scale, and straining as measured on a 5-point ordinal scale. Additional efficacy assessments based on weekly assessments include the Weekly Patient Assessment of Constipation Severity, the Weekly Patient Assessment of IBS Symptom Severity, the Weekly Patient Assessment of Adequate Relief, and the Weekly Patient Assessment of Treatment Satisfaction.

Safety Measures:

AE recording (each visit), clinical laboratory measures (chemistry and hematology: Screening, Randomization, Week 12/ETP, and Week 16/EOT Visits), body weight (each visit), vital sign parameters (each visit).

Statistical Methods:

Analysis Populations

- The Screened Population consists of all patients who had a Screening Visit (Visit 1) and were assigned a patient identification (PID) number.
- The Intent-to-Treat (ITT) Population consists of all randomized patients.
- The RW Population consists of all patients who were rerandomized or allocated to study drug upon completion of the Treatment Period.
- The Safety Population consists of all patients who received at least one dose of study drug.

General Methods

Efficacy outcomes from the Treatment Period will be based on the ITT Population and will be summarized by treatment group assigned at the Randomization Visit. Efficacy outcomes from the RW Period only or from the Combined Treatment and RW Period will be based on the RW Population and will be summarized by treatment sequence determined at the Week 12/ETP Visit. Treatment periods will be summarized for safety in the same manner as efficacy; however, safety summaries (Safety Population) will be based on actual treatment received.

Descriptive statistics including the number of patients, mean, minimum and maximum, and standard deviation (SD) will be calculated for continuous variables. Frequencies and percentages for each category will be calculated for categorical variables. Percentages will be based on the total number of non-missing values. The number missing will be presented, but without a percentage. All hypothesis tests will be two-sided at a 5% significance level, and 95% confidence intervals (CIs) will be used, unless stated otherwise.

Primary Efficacy Endpoint

- Change from Baseline in Abdominal Score at Each Week

  A patient's daily Abdominal Score is calculated as the average of the daily patient assessments of abdominal bloating at its worst, abdominal discomfort at its worst, and abdominal pain at its worst. If 2 or more of the individual daily abdominal symptoms are missing, then the Abdominal Score for that day will be missing. The weekly Abdominal Score is the average of the non-missing Abdominal Scores during each week (Weeks 1-12) in the trial. The baseline Abdominal Score is the average of the non-missing Abdominal Scores during the 14-day Pretreatment Period and the day of the Randomization Visit reported prior to randomization. Change from baseline will be calculated for each week as the weekly score minus the baseline score.

Primary Efficacy Analysis

Inferential testing between the linaclotide 290 µg and placebo groups with regard to the change from baseline in abdominal score over the Treatment Period will be evaluated employing a mixed model with repeated measures (MMRM) framework with week (categorical), treatment, geographic region, and week-by-treatment fixed effects, patient as the random effect, and baseline value as a covariate. An unstructured covariance structure will be utilized. Descriptive statistics based on the MMRM model will include least-squares (LS) mean change from baseline for each treatment, the LS mean difference between linaclotide 290 µg and placebo, corresponding 95% CIs, and the p-value associated with the comparison.

Secondary Efficacy Endpoints

- Change from Baseline in 12-week Abdominal Score (Cumulative Distribution Function [CDF])

  Daily Abdominal Score is calculated as described for the primary endpoint above. The 12-week Abdominal Score is the average on the non-missing Abdominal Scores reported over the course of the Treatment Period. The baseline Abdominal Score is the average of the non-missing Abdominal Scores during the 14-day Pretreatment Period and the day of the Randomization Visit reported prior to randomization. Change from baseline will be calculated as the 12-week score minus the baseline score.

- 6/12 Week Abdominal Score Responder

  A 6/12 Week Abdominal Score Responder is a patient who meets the Weekly Abdominal Score Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly Abdominal Score Responder is a patient who has an improvement from baseline of at least 2 points (ie, a -2 change from baseline) in the respective weekly abdominal score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly Abdominal Score Responder for that week.

Secondary Efficacy Analysis

The CDFs for linaclotide 290 µg and placebo will be estimated for the Change from Baseline in 12-week Abdominal Score endpoint. The CDFs will be compared using the Wilcoxon rank sum test with Hodges-Lehmann estimator for the median difference.

A secondary time-course analysis of the Change from Baseline in Abdominal Score (see primary endpoint definition above) will also be conducted. Using the MMRM framework defined in the primary analyses, treatment difference between linaclotide 290 µg and placebo will be assessed at each individual week. For each week, descriptive statistics based on the MMRM model will include the LS mean change from baseline for each treatment, the LS mean difference between linaclotide 290 µg and placebo, corresponding 95% CIs, and the p-value associated with the treatment comparison.

For the secondary responder endpoint, the proportion of responders in the linaclotide 290 µg group will be compared to the proportion of responders in the placebo group using a Cochran-Mantel-Haenszel (CMH) test controlling for geographic region. The number and percent of responders, the difference in responder rates between the linaclotide 290 µg and placebo groups, the odds ratio relative to placebo, all corresponding 95% CIs, and the p-value associated with the CMH test will be presented.

Controlling for Multiplicity

The overall family-wise Type I error rate for the primary and secondary efficacy analyses will be controlled at the α=0.05 level by employing a fixed-sequence procedure as described below. Following the fixed-sequence procedure, if the primary hypothesis between the placebo group and the linaclotide 290 µg group is statistically significant (α=0.05) in the ITT Population, then the primary objective of the trial will have been achieved and the next hypothesis can be tested; otherwise, testing will stop. The testing of each sequential hypothesis is conditional on all the previous hypotheses being rejected at the 0.05 level of significance. If a hypothesis is not rejected at the 0.05 level of significance, then all remaining hypotheses are deemed not statistically significant. All hypothesis testing will be two-sided.

1. Linaclotide vs. placebo – Change from Baseline in Abdominal Score (primary efficacy analysis)
2. Linaclotide vs. placebo – Change from Baseline in 12-week Abdominal Score (CDF) (secondary efficacy analysis)
3. Linaclotide vs. placebo – 6/12 Week Abdominal Score Responder (secondary efficacy analysis)
4. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 12
5. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 10
6. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 8
7. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 6
8. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 4
9. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 2
10. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 1

Additional Efficacy Endpoints

Additional efficacy endpoints will be explored outside of the formal testing procedures described above. These endpoints will include:

- 6/12 Week Abdominal Pain and Constipation (APC) +1 Responder
- Change from Baseline in Abdominal Pain at its Worst at Each Week
- Change from Baseline in CSBM Frequency Rate at Each Week
- Change from Baseline in SBM Frequency Rate at Each Week
- Change from Baseline in BSFS (Stool Consistency) at Each Week
- Change from Baseline in Stool Consistency at Each Week
- Change from Baseline in Straining at Each Week
- Change from Baseline in Abdominal Discomfort at its Worst at Each Week
- Change from Baseline in Abdominal Bloating at its Worst at Each Week
- Percent Change from Baseline in Abdominal Pain at its Worst at Each Week
- Change from Baseline in Percent of Days with Use of Rescue Medicine
- SBM Within 24 Hours After First Dose Responder
- 6/12 Week Abdominal Pain 30% Responder
  - ≥30% decrease from baseline in abdominal pain for ≥6 of 12 weeks
- 6/12 Week Abdominal Pain Responder
  - ≥2-point improvement from baseline in abdominal pain for ≥6 of 12 weeks
- 6/12 Week Abdominal Discomfort Responder
  - ≥2-point improvement from baseline in abdominal discomfort for ≥6 of 12 weeks
- 6/12 Week Abdominal Bloating Responder
  - ≥2-point improvement from baseline in abdominal bloating for ≥6 of 12 weeks
- 6/12 Week CSBM Responder
  - Increase of ≥1 CSBM/week from baseline for ≥6 of 12 weeks
- Treatment Satisfaction
- Adequate Relief
- 6/12 Week Adequate Relief Responder
- Change from Baseline in Constipation Severity

- Change from Baseline in IBS Symptom Severity

Additional Efficacy Analyses

Change-from-baseline endpoints and Treatment Satisfaction will be analyzed utilizing the same MMRM methods as described above for the primary analysis, and by visit as described for the secondary analyses. Responder endpoints will be analyzed using the methodology defined for the secondary responder analysis.

RW Period Analysis

All RW endpoints will be summarized for the RW Period only and for the Combined Treatment and RW Period using descriptive statistics and 95% CIs for each treatment sequence. No statistical inference will be performed.

Safety Analysis

All safety parameters will be analyzed descriptively in accordance with the General Methods.

Sample Size

The sample size of 600 patients (300 patients per treatment group) was chosen to ensure adequate power for testing the fixed-sequence procedure for the primary and secondary efficacy endpoints as defined above.

The power calculations for the primary endpoint are based on the placebo and linaclotide 290 μg treatment groups from the Phase 3 trial LIN-MD-31. The patients in LIN-MD-31 are considered representative of the patient population for this trial. Using a resampling with replacement-based simulation (1000 iterations) and controlling for multiplicity as outlined above, the trial will have >99% power to reject the primary endpoint and ~94% power to reject all primary and secondary hypotheses defined in the testing process.

Final Date: 26 March 2018

SCHEDULE OF EVALUATIONS

Schedule of Evaluations MCP-103-312							
Trial Period →	**Screening Period (Up to 21 days)**	**Pretreatment Period (14 to 21 days)**	**Treatment Period (12 weeks)**				**Randomized Withdrawal (RW) Period (4 weeks)[q]**
Visit →	**Screening Visit**	**Pretreatment Visit**	**Randomization Visit**	**Week 4 Visit**	**Week 8 Visit**	**Week 12/End-of-Treatment-Period Visit**	**Week 16/ End-of-Trial Visit [r]**
Visit Days →	**Day -42 through Day -15**	**Day -21 to Day -14**	**Day 1**	**Day 29±3**	**Day 57±3**	**Day 85+3**	**RW Day 29+3**
Visit Number →	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**
Trial Procedure ↓							
Signature of ICF	X						
Inclusion and Exclusion Criteria Verification	X	X	X*				
IWRS Registration [a]	X	X	X†	X	X	X	X
Medical History [b]	X						
Prior IBS-C Symptom Management Assessment [c]	X						
Physical Examination [d]	X					X*	X
Body Weight and Height [e]	X	X	X*	X	X	X*	X
Seated Vital Signs [f]	X	X	X*	X	X	X*	X
Prior and Concomitant Medicines [g]	X	X	X*	X	X	X*	X
Clinical Laboratory Tests [h]	X		X*			X*	X
Pregnancy Test [i]	X		X*			X*	X
Laxative/Suppository/ Enema Washout Instructions [j]	X						
AE Evaluations [k]		X	X*	X	X	X*	X
eDiary Registration and Training [l]		X	X*	X	X	X*	X
Daily and Weekly Assessments [m]		X	X*	X	X	X*	X

Schedule of Evaluations MCP-103-312							
Trial Period →	**Screening Period (Up to 21 days)**	**Pretreatment Period (14 to 21 days)**	**Treatment Period (12 weeks)**				**Randomized Withdrawal (RW) Period (4 weeks)[q]**
Visit →	**Screening Visit**	**Pretreatment Visit**	**Randomization Visit**	**Week 4 Visit**	**Week 8 Visit**	**Week 12/End-of-Treatment-Period Visit**	**Week 16/ End-of-Trial Visit [r]**
Visit Days →	**Day -42 through Day -15**	**Day -21 to Day -14**	**Day 1**	**Day 29±3**	**Day 57±3**	**Day 85+3**	**RW Day 29+3**
Visit Number →	**Visit 1**	**Visit 2**	**Visit 3**	**Visit 4**	**Visit 5**	**Visit 6**	**Visit 7**
Trial Procedure ↓							
eDiary Compliance Verification and Reminder [n]			X*	X	X	X*	X
Rescue Medicine Dispensed [o]		X	X	X	X	X	
Patient eDiary Entry, in Clinic			X*	X	X	X*	X
Randomization			X†			X	
IBS-QOL			X†	X	X	X	X
CDC HRQOL-4			X†			X	X
Study Drug Dispensed			X†	X	X	X[s]	
Study Drug Administration [p]			X				
Study Drug Accountability				X	X	X	X

AE=adverse event; BM=bowel movement; CBC=complete blood count; CDC HRQOL-4=Centers for Disease Control and Prevention Healthy Days Core Module; CIC=chronic idiopathic constipation; eDiary=electronic diary; EOT=End-of-Trial; ETP=End-of-Treatment-Period; IBS-C=irritable bowel syndrome with constipation; IBS-QOL=irritable bowel syndrome quality of life questionnaire; ICF=informed consent form; IWRS=interactive web response system; RW=randomized withdrawal.

* Done prior to randomization; † Done predose

a. Site personnel will interact with IWRS to register the patient visit. Refer to the IWRS User Manual.
b. Includes diagnosis using Rome IV criteria for IBS, which will be collected as part of the patient's disease-specific history. Rome IV diagnosis has no bearing on patient eligibility to participate in the trial (patients will be enrolled based Rome III criteria).
c. Prior IBS-C symptom management assessment includes prior treatments taken for IBS-C, lifestyle and diet modifications for alleviating symptoms of IBS-C, assessment of satisfaction with prior interventions' ability to relieve bowel and abdominal symptoms, and primary reason for stopping use.
d. A physical examination includes the following: general appearance, HEENT (head, ears, eyes, nose, and throat), neck, cardiovascular, thorax/lungs, breasts, abdomen, rectal, genitourinary, musculoskeletal, lymph nodes, skin, neurologic, and mental status. A rectal examination should be performed during the

Screening Period on all patients who do not require a colonoscopy (refer to Appendix 3). After the Screening Period, the rectal examination is optional and may be performed at the discretion of the investigator. Breast and genitourinary examinations are optional at the discretion of the investigator.

e. Height is measured only at the Screening Visit.
f. 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 five minutes.
g. 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 prior use of certain IBS-C or CIC (approved or unapproved) prescription medications (see Section 7.7 Concomitant Medicines).
h. Chemistry, CBC, and urine drug screen. The urine drug screen will be performed at the Screening Visit only.
i. To be eligible to continue in the trial, a negative serum pregnancy test must be documented at the Screening Visit. A negative urine pregnancy test must be documented at the Randomization Visit and the Week 12/ETP Visit (prior to randomization), and a negative serum pregnancy test must be documented at the Week 16/End-of-Trial (EOT) Visit.
j. Study coordinator will instruct patients about the use of laxatives, suppositories, and enemas (refer to Appendix 2).
k. All AEs occurring after the patient signs the ICF will be captured.
l. 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.
m. An eDiary will be used for recording Daily BM-related Symptom Severity Assessments, Daily Abdominal Symptom-severity Assessments, Weekly Assessments, and Use of Bisacodyl or Other Laxatives, Suppositories, or Enemas. Patients will enter BMs and laxative use in the eDiary on an event-driven basis, and will complete an evening entry each day to record daily abdominal assessments and any BMs and/or laxative use not previously recorded for that day.
n. 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 Week 16/EOT visit). eDiary questions may be found in the eDiary User Manual.
o. Rescue Medicine (oral bisacodyl or bisacodyl suppositories) will be supplied to patients at the Pretreatment Visit and, if needed, at subsequent study drug dispensing visits.
p. Study drug will be administered in the clinic at the Randomization Visit. Patients are instructed to fast for 2 hours before this clinic visit. On all other days, study drug will be taken once daily in the morning at least 30 minutes before breakfast.
q. The RW Period will start the day after the Week 12/ETP Visit (this will be considered RW Day 1).
r. Patients who are randomized but do not complete the Treatment Period or RW Period (withdraw consent or are discontinued before they have completed 12 weeks or 4 weeks of treatment, respectively), will be considered Treatment Period or RW Period withdrawals, respectively, and should complete the procedures required at the EOT Visit (even if out of window).
s. Patients will take their first dose of study drug in the RW Period on RW Day 1 (ie, the day after the Week 12/ETP Visit).

TRIAL IDENTIFICATION

A summary of key trial participants is provided in Table 1. All trial contact details will be provided prior to the Site Initiation Visit. Rather than contacting individuals listed within the table, sites should use the toll-free medical hotline for medical inquiries and the dedicated serious adverse event (SAE) fax/email for communication regarding SAEs, as listed below.

Table 1. Key Trial Participants

Role	Contact Information
Sponsor:	Ironwood Pharmaceuticals, Inc. 301 Binney Street Cambridge, MA 02142 617-621-7722 (Main Telephone) 617-494-0908 (Main Fax) www.ironwoodpharma.com
Sponsor Contact:	[redacted]
Sponsor Clinical Lead:	
Sponsor Medical Monitor:	
Sponsor Safety Officer:	
Clinical Laboratory:	
[redacted]	
[redacted]	

Role	Contact Information
Toll-free Medical Hotline Number:	1-800-201-8725
Dedicated Serious Adverse Event (SAE) Facsimile Number and Email:	Fax: 1-617-933-7688 clinicalsafety@ironwoodpharma.com

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LIST OF ABBREVIATIONS

AE	adverse event
ANCOVA	analysis of covariance
APC	Abdominal Pain and Constipation
BM	bowel movement
BMI	body mass index
BP	blood pressure
BSFS	Bristol Stool Form Scale
CFR	Code of Federal Regulations
CI	confidence interval
CIC	chronic idiopathic constipation
CBC	complete blood count
CDC HRQOL-4	Centers for Disease Control and Prevention Healthy Days Core Module
CDF	cumulative distribution function
CFTR	cystic fibrosis transmembrane conductance regulator
cGMP	cyclic guanosine monophosphate
CMH	Cochran-Mantel-Haenszel
CRO	contract research organization
CSBM	complete spontaneous bowel movement
DHHS	Department of Health and Human Services
DR	delayed release
eCRF	electronic case report form
EDC	electronic data capture
eDiary	electronic diary
EOT	End of Trial
ETP	End of Treatment Period
FDA	Food and Drug Administration
GC-C	guanylate cyclase-C

GCP	good clinical practice
GI	gastrointestinal
HEENT	head, ears, eyes, nose, and throat
HIPAA	Health Insurance Portability and Accountability Act
HT	hydroxytryptophan
IBD	inflammatory bowel disease
IBS	irritable bowel syndrome
IBS-C	irritable bowel syndrome with constipation
IBS-D	irritable bowel syndrome with diarrhea
IBS-M	irritable bowel syndrome mixed
IBS-U	unclassified irritable bowel syndrome
ICF	informed consent form
ICH	International Conference on Harmonisation
IR	immediate release
IRB	Institutional Review Board
ITT	intent-to-treat
IWRS	Interactive web response system
LAR	legally authorized representative
MedDRA	Medical Dictionary for Regulatory Activities
MMRM	mixed model with repeated measures
NDA	New Drug Application
NEAE	newly-emergent adverse event
NRS	numerical rating scale
PCS	potentially clinically significant
PID	patient identification
QOL	quality of life
RW	Randomized Withdrawal
SAE	serious adverse event

SAP	statistical analysis plan
SBM	spontaneous bowel movement
SD	standard deviation
SE	standard error
SOC	system organ class
TEAE	treatment-emergent adverse event
US	United States

1. ETHICAL CONSIDERATIONS

This clinical trial is designed to comply with the International Conference on Harmonisation (ICH) Guidance on General Considerations for Clinical Trials, ICH E8 (published in the US Federal Register Volume 62, page 66113, December 17, 1997) and Good Clinical Practice (GCP): Consolidated Guidance, ICH E6 (published in the US Federal Register, Volume 62, page 25692, May 9, 1997). The trial will be conducted in full compliance with the US Food and Drug Administration (FDA) guidelines for GCP and in accordance with the ethical principles that have their origins in the Declaration of Helsinki.

1.1 INSTITUTIONAL REVIEW BOARD

Obtaining approval by the Institutional Review Board (IRB) prior to the start of the trial will be the responsibility of the investigator. A copy of the approval letter will be transmitted to the Sponsor or designee. During the course of the trial, the investigator will provide timely and accurate reports to the IRB on the progress of the trial, and will notify the IRB of serious adverse events (SAEs) or other significant safety findings in a manner consistent with IRB policies and ICH and GCP requirements. The trial protocol (and any amendments), Informed Consent Form (ICF; Appendix 1), and associated documentation will be approved by the IRB prior to trial initiation, in compliance with 21 CFR Part 56.

1.2 PATIENT INFORMATION AND CONSENT

Before entry into the trial, patients will be provided with a written explanation of the trial describing the nature of the study, as well as its purpose, expected duration, and the benefits and risks involved in trial participation per 21 CFR Part 50. Patients will then be given the opportunity to ask questions and will be informed of their right to withdraw from the trial without prejudice. After this explanation and before entering the trial, the patient will voluntarily sign an ICF.

If new information becomes available that may be relevant to the patient's consent and willingness to participate in the trial, the ICF will be revised and any currently enrolled patient will be made aware of the new information and asked if he/she wishes to continue in the trial. The revised consent form will be submitted to the IRB for review and approval prior to its use.

2. INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

This trial will be performed at approximately 80 trial centers in the US. The investigator at the trial center will be responsible for ensuring that the trial is conducted according to the signed Clinical Trial Agreement, the protocol, IRB requirements, and GCP guidelines.

The investigator will be responsible for the oversight of the site's conduct of the trial, which will consist of completing all protocol assessments, maintaining the trial file and the patient records, drug accountability, corresponding with the IRB, and completing the electronic case report forms (eCRFs).

Ironwood Pharmaceuticals, Inc. and Allergan, Plc are development partners for this trial.

3. INTRODUCTION

3.1 IBS-C

Irritable bowel syndrome (IBS) is a chronic functional gastrointestinal (GI) disorder characterized by recurrent symptoms of abdominal pain and/or discomfort, accompanied by altered bowel function and a feeling of bloating.(2-6) In moderate to severe cases of IBS, an overall deterioration in quality of life (QOL) is often present.(7) IBS is one of the most frequently seen disorders in the United States; data suggest the prevalence of IBS is 11-14% of the adult population.(7) IBS is subtyped as IBS with diarrhea (IBS-D) or IBS with constipation (IBS-C), based on Rome criteria. Patients who meet criteria for both diarrhea and constipation are subtyped as IBS mixed (IBS-M); patients who do not fit into any of these three subtypes are classified as having unclassified IBS (IBS-U).(8)

3.2 LINACLOTIDE

Linaclotide (LINZESS®) is an orally-administered, minimally-absorbed 14-amino acid synthetic peptide agonist of guanylate cyclase C (GC-C) approved by FDA in 2012 for the treatment of IBS-C and chronic idiopathic constipation (CIC) in adults. As a GC-C agonist, linaclotide and its active metabolite improve bowel symptoms and abdominal pain via agonism of cyclic guanosine monophosphate (cGMP) on the intestinal mucosa. Linaclotide induces fluid secretion in two ways: 1) by increasing intracellular cGMP leading to activation of cystic fibrosis transmembrane conductance regulator (CFTR) ion channels, resulting in secretion of chloride and bicarbonate into the intestinal lumen, resulting in accelerated transit;(9) and 2) by inhibiting sodium reabsorption in the colon.(10) These effects on fluid dynamics within the GI tract are believed to play a role in accelerating intestinal transit and increasing stool water, which in turn results in softer stools. Orally administered linaclotide has also been shown to reduce visceral hypersensitivity in animal models and abdominal pain in patients with IBS-C.(11) Linaclotide is believed to reduce abdominal pain by acting locally in the intestinal mucosa to reduce firing of mechanosensitive afferent nerve fibers.

3.3 LINACLOTIDE CLINICAL DEVELOPMENT

The clinical development program for LINZESS® that culminated in FDA approval for use in adult patients with IBS-C included two large double-blind, placebo-controlled registration trials.(12, 13) These trials evaluated the safety and efficacy of linaclotide at a dose of 290 µg, administered as an oral capsule. One trial had a 26-week Treatment Period and one trial had a 12-week Treatment Period followed by a 4-week Randomized Withdrawal Period; however, the overall design of both Phase 3 trials was identical through the first 12 weeks of the Treatment Period. Both trials met all pre-specified primary and secondary efficacy endpoints. For each of the four primary efficacy parameters, the linaclotide dose group in both trials had statistically significantly higher proportions of responders compared with placebo (controlling for multiplicity). For all secondary efficacy change-from-baseline parameters (including complete spontaneous bowel movement [CSBM] frequency rate, spontaneous bowel movement [SBM] frequency rate, stool consistency, straining, abdominal bloating, abdominal discomfort, abdominal pain, and abdominal pain-free days) and responder parameters (6/12 Week CSBM +1 Responder and 6/12 Week Abdominal Pain Responder), the linaclotide group demonstrated statistically significant improvement compared with the placebo group ($p<0.01$ for all secondary parameters controlling for multiplicity) in both trials.

Linaclotide was well-tolerated in these trials.(12, 13) There were no deaths. One patient died during the Screening Period but never received either linaclotide or placebo. There were a total of 15 SAEs in the 2 trials. There were 9 (1.1%) SAEs in placebo-treated patients and 6 (0.7%) SAEs in linaclotide-treated patients. Overall, there was no obvious pattern in the types of SAEs experienced in either the placebo or linaclotide group. There were no SAEs of diarrhea. Treatment-emergent AEs (TEAEs) occurred in 54.9% of placebo-treated and 60.8% of linaclotide-treated patients. Diarrhea was the most frequent TEAE and was reported in 3.0% of patients treated with placebo and 19.8% of patients treated with linaclotide. Diarrhea was generally mild to moderate in severity. Diarrhea resulted in the discontinuation of 0.4% of patients treated with placebo and 5.3% treated with linaclotide.

More recently, a Phase 2b, multicenter, randomized, double-blind, double-dummy, placebo-controlled, parallel-group, dose-range-finding study evaluated 2 different delayed release (DR) tablet formulations of linaclotide (DR1 and DR2), the FDA-approved 290-µg LINZESS® (the immediate release [IR] capsule formulation of linaclotide), and placebo administered once daily for 12 weeks. A total of 532 patients with IBS-C (Rome III criteria) were randomized in equal proportions to 1 of 8 treatment groups:

- Linaclotide DR1 (30 µg, 100 µg, 300 µg)
- Linaclotide DR2 (30 µg, 100 µg, 300 µg)
- Linaclotide IR (LINZESS®) 290 µg
- Placebo

All 532 patients randomized to treatment (ITT Population) took at least 1 dose of study drug (Safety Population). Demographics and baseline clinical characteristics were generally similar across the 8 treatment groups, which each included 66-67 patients. Results for the linaclotide IR 290 µg and placebo groups are summarized below.

The key efficacy endpoints were: change from baseline (CFB) in weekly abdominal pain (-1.9 in the linaclotide IR group versus -1.4 in the placebo group), CFB in weekly CSBM frequency rate (2.1 in the linaclotide IR group versus 1.1 in the placebo group; $p<0.05$), and 6/12 Week Abdominal Pain and Constipation (APC) +1 Responder (31.8% in the linaclotide IR group versus 21.2% in the placebo group). Improvements from baseline in the individual abdominal symptoms (abdominal bloating, abdominal discomfort, and abdominal pain) and in the abdominal score (a multi-component score based on these individual abdominal symptoms) were seen in the linaclotide IR group compared with the placebo group.

Linaclotide IR was well-tolerated in this study and safety results were consistent with the known safety profile of linaclotide. There were no deaths in the study. Two patients reported on-treatment SAEs: 1 placebo patient (pneumonia and sepsis) and 1 linaclotide IR patient (gastroenteritis); all 3 SAEs were considered unrelated to study drug by the investigator. As in previous linaclotide trials, the most common TEAE was diarrhea (13.6% in the linaclotide IR group versus 1.5% in the placebo group). All diarrhea events were mild or moderate in severity. Diarrhea resulted in the discontinuation of 6.1% of patients treated with linaclotide IR and 0% of patients treated with placebo.

Refer to the Investigator's Brochure for a more detailed description of the chemistry, pharmacology, efficacy, and safety of linaclotide, based on studies conducted in animals, healthy volunteers, and in patients with IBS-C and CIC.

4. TRIAL OBJECTIVES

The objective of this trial is to evaluate the efficacy on abdominal symptoms (abdominal bloating, abdominal discomfort, and abdominal pain) and safety of linaclotide 290 µg administered orally to patients with irritable bowel syndrome with constipation (IBS-C).

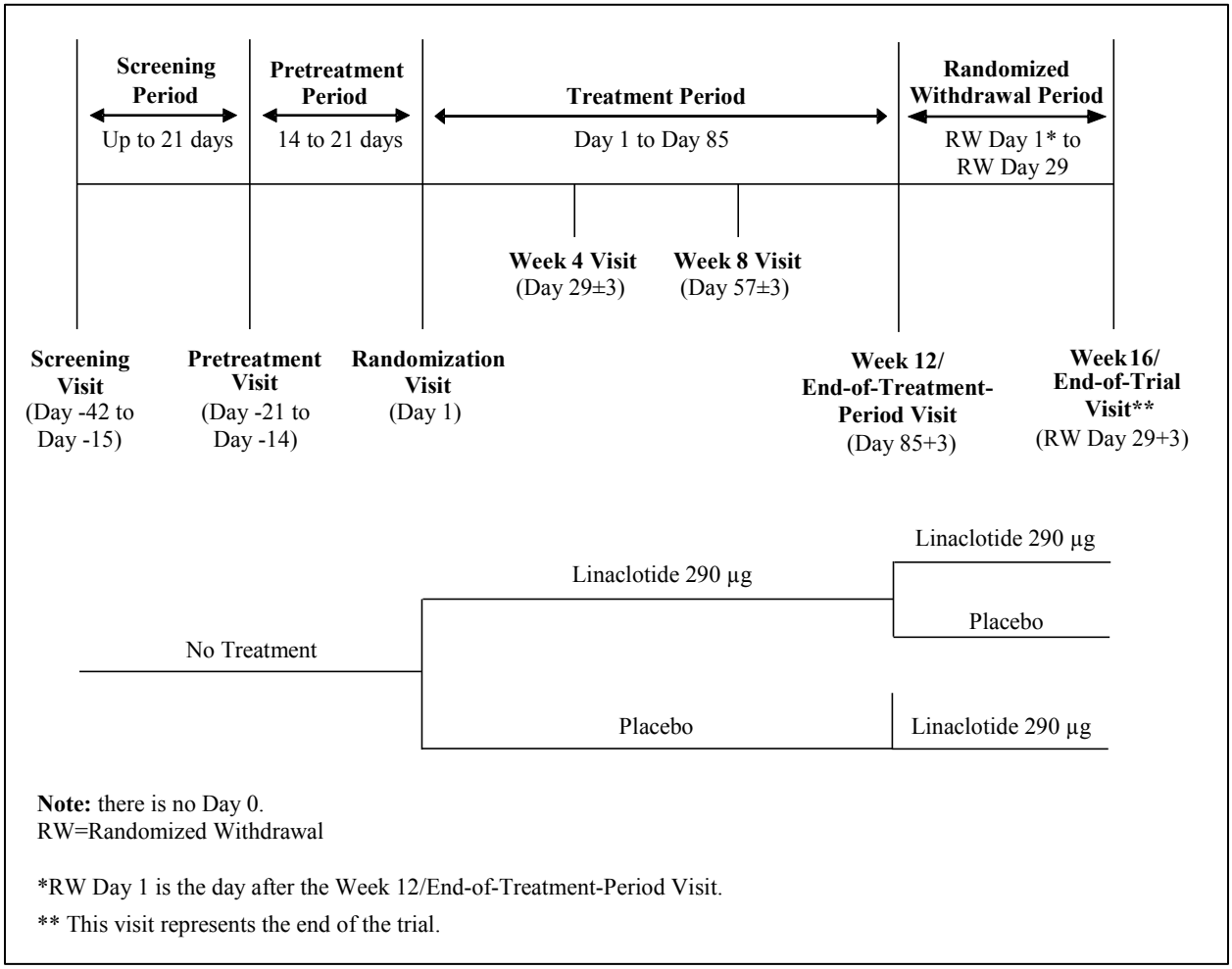
5. INVESTIGATIONAL PLAN

5.1 OVERALL TRIAL DESIGN AND PLAN

This is a multicenter, randomized, double-blind, placebo-controlled, parallel-group trial, consisting of 4 distinct periods, as illustrated in Figure 1 below. The trial will enroll patients who have IBS-C diagnosed using Rome III criteria. Eligible patients will be randomized to 1 of 2 treatments: linaclotide 290 µg or placebo (1:1) once daily. Patients who complete the 12-week Treatment Period will enter the 4-week Randomized Withdrawal (RW) Period and be allocated to study drug in a double-blind manner, as follows:

- Patients randomized to linaclotide 290 µg during the Treatment Period will be rerandomized to linaclotide 290 µg or placebo (1:1)
- Patients randomized to placebo during the Treatment Period will be allocated to linaclotide 290 µg

Figure 1. Overview of Trial Design



Note: there is no Day 0.
RW=Randomized Withdrawal

*RW Day 1 is the day after the Week 12/End-of-Treatment-Period Visit.

** This visit represents the end of the trial.

5.1.1 Trial Periods

5.1.1.1 Screening Period

The Screening Period starts with the signing of the informed consent form (ICF; Appendix 1) and may last for up to 21 days. During this period, patient eligibility for entry into the Pretreatment Period will be determined. 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 (see Appendix 2), the Screening Visit and Pretreatment Visit may be combined into one visit.

5.1.1.2 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 Bowel Movement (BM)-related Symptom-severity Assessments on an event-driven basis (meaning these are assessments made at the time the event occurs)
- Daily Abdominal Symptom-severity Assessments in an evening report
- Weekly Patient Assessment of Constipation Severity
- Weekly Patient Assessment of IBS Symptom Severity
- Weekly Patient Assessment of Adequate Relief
- Use of Bisacodyl or Other Laxatives, Suppositories, or Enemas on an event-driven basis

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

5.1.1.3 Treatment Period

The Treatment Period begins with treatment assignment and lasts for 12 weeks. Patients will be randomized to 1 of 2 treatments: linaclotide 290 µg or placebo (1:1). Patients will take their initial dose of study drug at the trial center during the Randomization Visit, after fasting for 2 hours. On all other days, study drug will be taken once daily in the morning at least 30 minutes before breakfast. Patients will continue to use the handheld eDiary to provide their Daily BM-related and Abdominal Symptom-severity Assessments, weekly assessments (as described above and including Weekly Patient Assessment of Treatment Satisfaction), and Use of Bisacodyl or Other Laxatives, Suppositories, or Enemas. Other patient-reported outcomes assessments (IBS Quality of Life Questionnaire [IBS-QOL] and CDC Healthy Days Core Module [CDC HRQOL-4]) will be administered at trial visits throughout the Treatment Period. Patients will complete a Week 4 Visit, Week 8 Visit, and Week 12/End-of-Treatment-Period (ETP) Visit during the Treatment Period (see Schedule of Evaluations).

5.1.1.4 Randomized Withdrawal (RW) Period

The RW Period is defined as the 4 weeks immediately following the Treatment Period. Patients who complete the 12-week Treatment Period and return for the Week 12/ETP Visit will enter the 4-week RW Period and be allocated to study drug in a double-blind manner, as follows:

- Patients randomized to linaclotide 290 μg during the Treatment Period will be rerandomized to linaclotide 290 μg or placebo (1:1)
- Patients randomized to placebo during the Treatment Period will be allocated to linaclotide 290 μg

Patients will take their first dose of study drug in the RW Period the day after the Week 12/ETP Visit (this will be considered RW Day 1). As during the Treatment Period, study drug will be taken once daily in the morning at least 30 minutes before breakfast. Patients will continue to use the handheld eDiary to provide their daily assessments, weekly assessments, and laxative use. Other patient-reported outcomes will be administered at the trial visit during the RW Period. At the end of the RW Period, patients will complete the Week 16/End-of-Trial (EOT) Visit.

5.2 RATIONALE FOR TRIAL DESIGN AND CONTROL GROUP

A double-blind, placebo-controlled, parallel-group trial 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 trial has a 14-21 day Pretreatment Period to establish a baseline without therapy 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 4-week RW Period to determine if a rebound (or worsening of symptoms from baseline) or other withdrawal effects might occur after linaclotide treatment has been withdrawn.

5.3 TRIAL DURATION

The test product and reference therapy will be administered for a total of 16 weeks. Total patient participation is expected to last for up to 162 days.

6. SELECTION OF TRIAL POPULATION

6.1 INCLUSION CRITERIA

To be eligible to participate in the trial, patients must meet the following criteria:

1. Patient has given consent by signing an ICF.
2. Patient is ambulatory (females must also be non-pregnant) and is aged 18 years or older at the Screening Visit. Lactating females must agree not to breastfeed.
3. Female patients of childbearing potential (ie, women who are not postmenopausal or who have not had a bilateral oophorectomy, hysterectomy, or tubal ligation) who are sexually active with a male partner must agree to use one of the following methods of birth control from the date they sign the ICF until 24 hours after their final dose of study drug:
   a. Hormonal contraception (ie, oral contraceptive, contraceptive implant, or injectable hormonal contraceptive)
   b. Double-barrier birth control (eg, condom plus intrauterine device, diaphragm plus spermicide)
   c. Maintenance of a monogamous relationship with a male partner who has been surgically sterilized by vasectomy
4. Females of childbearing potential must have a negative serum pregnancy test at the Screening Visit and a negative urine pregnancy test at the Randomization Visit prior to dosing.
5. Patient meets the colonoscopy requirements as defined by the American Gastroenterological Association guidelines and described in Appendix 3. (Note: Patients who receive narcotic anesthesia for a colonoscopy are eligible to enter the Pretreatment Period on the fifth day after the colonoscopy.)
6. Patient meets Rome III criteria for IBS (1): reports abdominal pain or discomfort at least 3 days/month during the 3 months before the diagnosis with the onset at least 6 months before the diagnosis, associated with two or more of the following features:
   a. Improved with defecation
   b. Onset associated with a change in frequency of stool
   c. Onset associated with a change in form (appearance) of stool

   Note: the diagnosis can be made at the Screening Visit or can be based on symptoms that the patient had before starting chronic treatment with linaclotide (LINZESS®), lubiprostone (AMITIZA®), or plecanatide (TRULANCE™).

7. During the time when the Rome III criteria for IBS were met, the patient had hard or lumpy stools (Bristol Stool Form Scale [BSFS] scores of 1 or 2) with at least 25% of BMs and had loose (mushy) or watery stools (BSFS scores of 6 or 7) with less than 25% of BMs in the absence of antidiarrheal or laxative use.
8. Patient reports <3 BMs (with each BM occurring in the absence of any laxative, suppository, or enema use during the preceding 24 hours) per week for at least 12 weeks, before the Screening Visit or before starting chronic treatment with linaclotide (LINZESS®), lubiprostone (AMITIZA®), polyethylene glycol 3350 (MiraLAX®), plecanatide (TRULANCE™), or any laxative.
9. Patient has an average score for abdominal pain at its worst of ≥3.0, as reported in the eDiary using an 11-point numerical rating scale (NRS) during the 14 days before the Randomization Visit and including the data entry into the eDiary made in clinic at the Randomization Visit prior to randomization.
10. Patient reports ≤6 complete spontaneous BMs (CSBMs) and ≤10 spontaneous BMs (SBMs) in the eDiary occurring over the 14 days before the Randomization Visit and including the data entry into the eDiary made in clinic at the Randomization Visit prior to randomization. (Note: A CSBM is an SBM that is associated with a sense of complete evacuation. An SBM is a BM that occurs in the absence of laxative, suppository, or enema use on the calendar day of the BM or the calendar day before the BM as evidenced by the patient-reported laxative use via the eDiary.)
11. Patient is compliant (as defined in the trial procedures section below) with eDiary completion by adequately responding to eDiary questions (ie, completing the daily evening report) on 10 or more of the 14 days before the Randomization Visit.
12. Patient is willing to discontinue any laxatives used before the Pretreatment Visit in favor of the protocol-defined Rescue Medicine (bisacodyl tablets or suppositories).
13. Patient is fluent in English or Spanish.
14. Patient agrees to refrain from making any new, major life-style changes that may affect IBS-C symptoms (eg, starting a new diet or changing his or her exercise pattern) from the time of signature of the ICF to the last trial visit.

6.2 EXCLUSION CRITERIA

Patients who meet any of the following criteria will not be eligible to participate in the trial.

1. Patient reports loose (mushy) or watery stools (BSFS score of 6 or 7) in the absence of any laxative, suppository, enema, or prohibited medicine (as described in Appendix 2) for >25% of BMs during the 12 weeks before the Screening Visit.

2. Patient has clinically significant concurrent illness or findings on a physical examination or clinical laboratory tests (clinical chemistry panel, complete blood count [CBC], urine drug screen) after signing the ICF but before receiving the first dose of study drug. (Note: The investigator will determine if a particular finding is clinically significant. In making this determination, the investigator will consider whether the particular finding could prevent the patient from performing any of the protocol-specified assessments, could represent a condition that would exclude the patient from the trial, could represent a safety concern if the patient participates in the trial, or could confound the trial-specified assessments of safety or efficacy.)
3. Patient has been diagnosed with or has a family history of familial adenomatous polyposis, hereditary nonpolyposis colorectal cancer, or any other form of familial colorectal cancer.
4. Patient has a prior history of inflammatory bowel disease (IBD).
5. Patient currently has clinically significant symptoms such as lower GI bleeding (rectal bleeding or heme-positive stool), iron-deficiency anemia, weight loss, or systemic signs of infection or colitis.
6. Patient currently has active peptic ulcer disease (ie, not adequately treated or stable with therapy).
7. Patient has a history of diverticulitis or any chronic condition (eg, chronic pancreatitis, polycystic kidney disease, ovarian cysts, endometriosis) with ongoing symptoms that can be associated with abdominal pain or discomfort and could confound the assessments in this trial.
8. Patient has a central nervous system cause of constipation (eg, Parkinson's disease, spinal cord injury, and multiple sclerosis).
9. Patient has ever had any of the following diseases or conditions that can be associated with constipation: pseudo-obstruction, megacolon, megarectum, bowel obstruction, descending perineum syndrome, solitary rectal ulcer syndrome, systemic sclerosis.
10. Patient has ever had a fecal impaction that required hospitalization or emergency room treatment, or has a history of cathartic colon, laxative or enema abuse, ischemic colitis, or pelvic floor dysfunction (unless successful treatment has been documented by a normal balloon expulsion test).
11. Patient has a known or suspected structural abnormality of the gastrointestinal (GI) tract (eg, mechanical gastrointestinal obstruction) or a disease or condition that can affect GI motility.
12. Patient has a history of hypersensitivity to linaclotide or to any of the excipients contained in the study drug (active or placebo) as described in Section 7.1.
13. Patient has had surgery that meets any of the following criteria:
    a. Bariatric surgery for treatment of obesity, or surgery to remove a segment of the GI tract at any time before the Screening Visit

b. Surgery of the abdomen, pelvis, or retroperitoneal structures during the 6 months before the Screening Visit
c. An appendectomy or cholecystectomy during the 60 days before the Screening Visit
d. Other major surgery during the 30 days before the Screening Visit

14. Patient has a history of cancer other than treated basal cell or squamous cell carcinoma of the skin. (Note: Patients with a history of cancer are allowed if the malignancy has been in a complete remission for at least 5 years before the Randomization Visit. A complete remission is defined as the disappearance of all signs of cancer in response to treatment.)
15. Patient has a history of diabetic neuropathy.
16. Patient has untreated hypothyroidism or treated hypothyroidism for which the dose of thyroid hormone has not been stable for at least 6 weeks at the time of the Screening Visit.
17. Patient has a recent history (during the 12 months before the Randomization Visit) of drug or alcohol abuse. (Note: Patients with a history of drug or alcohol abuse that was diagnosed greater than 12 months before the Randomization Visit may be enrolled if they have exhibited no actual abuse during the 12 months before the Randomization Visit.)
18. Patient reports a BSFS score of 6 (loose, mushy stools) for >1 SBM or a BSFS score of 7 (watery stools) with any SBM occurring over the 14 days before the Randomization Visit and including the data entry into the eDiary made in the clinic at the Randomization Visit prior to randomization.
19. Patient used Rescue Medicine (bisacodyl tablet or suppository) or any other laxative, suppository, or enema, on the day before or the day of the Randomization Visit prior to randomization.
20. Patient reported using a Prohibited Medicine (excluding laxatives, suppositories, and enemas) during the Pretreatment Period or is not willing or able to abide by the restrictions regarding use of Prohibited Medicines defined in Appendix 2. (Note: The use of fiber, bulk laxatives, or stool softeners [such as docusate] is acceptable provided the patient has been on a stable dose during the 30 days before the Screening Visit and plans to continue on a stable dose throughout the trial.)
21. Patient has been hospitalized for a psychiatric condition or has made a suicide attempt during the two years before the Randomization Visit.
22. Patient has taken commercially available linaclotide (LINZESS®) or plecanatide (TRULANCE™) or received an investigational drug during the 30 days before the Screening Visit.
23. Patient has an acute or chronic condition that, in the investigator's opinion, would limit the patient's ability to complete or participate in this clinical trial.
24. Patient has previously entered the Pretreatment Period of this trial.

25. Patient is directly or indirectly involved in the conduct and administration of this trial as an investigator, sub-investigator, study coordinator, other trial staff member, or employee of Ironwood Pharmaceuticals, Forest Laboratories, Actavis, or Allergan; or the patient is a first-degree family member, significant other, or relative residing with one of the above persons involved directly or indirectly in the trial; or the patient is enrolled in this trial at another clinical trial site.

6.3 DISCONTINUATION OF PATIENTS FROM THERAPY OR ASSESSMENT

A premature discontinuation will occur when a patient who signed the ICF ceases participation in the trial, regardless of circumstances, before the completion of the protocol. Patients can prematurely discontinue or be prematurely discontinued by the investigator or Sponsor from the trial at any time for any reason including the following:

- Failure to meet Inclusion/Exclusion Criteria (Screening or Pretreatment Failure)
- Adverse event (AE)
- Lack of efficacy
- Protocol violation
- Non-compliance with study drug
- Withdrawal of consent
- Lost to follow-up (every effort must be made to contact the patient; a certified letter must be sent)
- Trial termination by Sponsor
- Pregnancy
- Other, such as administrative reasons

Patients who are randomized but do not complete the Treatment Period or RW Period (withdraw consent or are discontinued before they have completed 12 weeks or 4 weeks of treatment, respectively) will be considered Treatment Period or RW Period withdrawals, respectively, and should complete the procedures required at the Week 16/EOT Visit (even if out of window) at the time of their discontinuation. The reasons for premature discontinuation from the trial will be documented on the study termination form of the eCRF. All data on the patient prior to discontinuation will be made available to Ironwood (or designee). Any patient who withdraws because of an AE will be followed until the AE resolves, stabilizes, or can be explained as being unrelated to study drug. The trial centers should make a reasonable effort to follow pregnant patients until delivery or end of the pregnancy.

If a patient does not return for a scheduled termination visit (EOT Visit), the trial center should contact the patient. Every effort is to be made to contact the patient, including sending a certified letter. In every case, the patient outcome, including lost to follow-up information, will be documented.

The investigator may allow a patient to stop taking study drug for up to 3 days should an intolerable AE occur. If the investigator believes that the patient is unable to resume dosing or requires a suspension of dosing on more than one occasion, the investigator should contact the Medical Monitor to discuss the patient's continued participation in the trial.

6.4 REPLACEMENT PROCEDURES

Patients in this trial who prematurely discontinue treatment will not be replaced.

7. TREATMENTS

7.1 TREATMENTS ADMINISTERED

Study drug in the form of oral capsules will be provided by Ironwood or designee. For the double-blind Treatment Period and RW Period, patients will be supplied with identically appearing capsules containing linaclotide 290 µg or placebo.

Linaclotide will be provided as 290-µg oral capsules containing active pharmaceutical ingredient with the following excipients: microcrystalline cellulose, calcium chloride dihydrate, l-leucine, hypromellose, and gelatin.

Matching placebo will be provided as oral capsules containing microcrystalline cellulose and gelatin.

7.2 IDENTITY AND STORAGE OF INVESTIGATIONAL PRODUCT

All study drug will be supplied in bottles containing thirty-five (35) capsules of linaclotide 290 µg or matching placebo capsules. Bottles will have child-resistant screw-top caps and will be uniquely numbered and labeled in a double-blind fashion that conforms to regulatory requirements.

All study drug will be provided by Ironwood or designee. Linaclotide 290 µg and matching placebo capsules will be stored at the trial center in an appropriate secure, temperature-controlled area at 77°F (25°C), excursions permitted to 59-86°F (15-30°C). Any deviations from the storage conditions must be reported to Ironwood and use of the study medication suspended until authorization for its continued use has been provided by Ironwood.

7.3 METHOD OF ASSIGNING PATIENTS TO TREATMENT GROUPS

The patient identification (PID) number will consist of 7 digits; [redacted] The patient will retain the same PID number (which is also the Screening Number) throughout the Treatment Period.

Patients who meet all of the inclusion criteria and none of the exclusion criteria will be randomized into the trial at the Randomization Visit on Day 1. Approximately 600 patients will be randomized to 1 of 2 treatments: linaclotide 290 µg or placebo (1:1). Patients who complete the Treatment Period will enter the RW Period and be allocated to study drug as follows.

- Patients randomized to linaclotide 290 µg during the Treatment Period will be rerandomized to linaclotide 290 µg or placebo (1:1)
- Patients randomized to placebo during the Treatment Period will be allocated to linaclotide 290 µg

Randomization numbers will be assigned by an interactive web response system (IWRS).

7.4 SELECTION OF DOSAGE IN THE STUDY

The 290-µg dose of linaclotide was selected for this Phase 3b trial to further evaluate the efficacy of the FDA-approved dose of linaclotide on abdominal symptoms (abdominal bloating, abdominal discomfort, and abdominal pain) in IBS-C patients.

7.5 SELECTION AND TIMING OF DOSE FOR EACH PATIENT

All study drug will be administered orally once daily. Patients who meet all eligibility criteria at the Screening and Pretreatment Visits will be randomized to treatment (as described in Section 7.3) at the Randomization Visit (randomization number assigned by IWRS) and dispensed one bottle containing 35 tablets. Additional bottles will be dispensed at subsequent visits per the Schedule of Evaluations. Patients will be instructed to take one capsule in the morning at least 30 minutes before breakfast. Patients will take their initial dose of study drug (one capsule) at the trial center during the Randomization Visit; patients must have fasted for at least 2 hours prior to arriving at the clinic for this visit. Patients will take study drug as usual in the morning before the Week 12/ETP Visit. Patients will be instructed to return all unused study drug and bottles to the study center at the visits defined in the Schedule of Evaluations.

At the Week 12/ETP Visit, patients who have not withdrawn from the Treatment Period will be rerandomized by the IWRS (as described in Section 7.3) and dispensed a bottle containing 35 tablets of double-blind study drug for the RW Period. Treatment will be taken according to the dosing instructions for the Treatment Period. Patients will take their initial dose of study drug in the RW Period (1 tablet) on RW Day 1 (ie, the day after the Week 12/ETP Visit).

7.6 BLINDING

The Sponsor study personnel, the investigator and all other site study personnel, and the patient will remain blinded to individual patient treatment assignments throughout the trial. Specific designated personnel in the Drug Safety and Pharmacovigilance group at Ironwood may be unblinded to the treatment assignment of individual patient for regulatory reporting purposes.

Patient randomization codes for the Treatment Period and for the RW Period will be generated by Allergan and implemented by the IWRS vendor (an electronic version will be stored on a secure server). The Treatment Period randomization list will identify each patient by randomization number and include the patient's corresponding treatment assignment. The RW Period list will be stratified by the treatment assigned in the Treatment Period and will include the stratum, patient's rerandomization number, and the corresponding treatment assignment. The medication code list will be supplied by Almac Clinical Services 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 study drug for the welfare of the patient, and only after unsuccessfully attempting to contact the Medical Monitor or designee. If the blind is broken, the trial center will notify the Ironwood contact (see the Study Identification information in the Synopsis) immediately. An explanation for breaking the blind will be recorded on the relevant eCRF. Breaking the code at the trial center will disqualify the patient from further participation in the trial (refer to Section 6.3 for details regarding procedures and follow-up for patients who are discontinued from therapy).

7.7 CONCOMITANT MEDICINES

A complete list of drugs that are conditionally allowed and drugs that are not allowed as concomitant medicines for either episodic or chronic use or as Rescue Medicine is provided in Appendix 2. During the Pretreatment, Treatment, and RW Periods, patients may use dispensed, protocol-permitted laxatives (bisacodyl tablets or suppositories) as Rescue Medicine when at least 72 hours have passed since their previous BM or when their symptoms become intolerable. In order to qualify for randomization into the Treatment Period, patients must not have used Rescue Medicine on the day before the Randomization Visit or on the day of the Randomization Visit prior to randomization. Patients must agree to refrain from using Rescue Medicine from the time they arrive at the clinic for the Randomization Visit through the day after randomization.

At the Screening Visit, all ongoing medicines or investigational products taken by the patient will be recorded. Past use of certain medications to manage IBS-C symptoms, even if not ongoing at the time of the Screening Visit, will be recorded in the Prior IBS-C Symptom Management Assessment (refer to Section 8.2.6 and Appendix 7) at the Screening Visit.

Any over-the-counter or prescription laxatives, suppositories, or enemas used to treat IBS-C may not be used during the Pretreatment, Treatment, and RW Periods and may not be used on the day before the Pretreatment Visit. Other prohibited medicines may not be used during the Pretreatment, Treatment, and RW Periods and may not be used during the 14 days before the Pretreatment Visit (refer to Appendix 2). Thereafter, any changes in concomitant medicines or new medicines added will be recorded on the eCRF. Concomitant medicines will be recorded at study visits throughout the entire trial. Rescue Medicine use will be documented by the patient via eDiary.

7.8 TREATMENT COMPLIANCE

Study drug will be administered to the patient by study center staff at the Randomization Visit. For all other days in the Treatment Period and RW Period, study drug will be taken by the patient once daily in the morning at least 30 minutes before breakfast. Study drug accountability will be closely monitored during the Treatment Period and RW Period by counting the number of capsules returned and recording that information on the eCRF. Every effort will be made to collect all unused study drug. Additionally, patients will be asked about the number of capsules lost and that information will be recorded on the eCRF.

8. STUDY PROCEDURES AND ASSESSMENTS

8.1 EFFICACY ASSESSMENTS

8.1.1 Primary Efficacy Assessments

The efficacy assessments that will be used to determine the primary efficacy endpoint (Section 10.4.1) are the daily patient assessments of abdominal pain, abdominal discomfort, and abdominal bloating at their worst (ie, the components of the abdominal score).

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 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">

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 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">

8.1.2 Secondary Efficacy Assessments

The efficacy assessments that will be used to determine the secondary efficacy endpoints are the daily patient assessments of abdominal pain, abdominal discomfort, and abdominal bloating at their worst, as described in Section 8.1.1.

8.1.3 Additional Efficacy Assessments

In addition to the primary/secondary efficacy assessments, the following efficacy assessments are used in determining the additional efficacy endpoints.

Complete Spontaneous Bowel Movement

The CSBM assessment is based on the eDiary questions that determine whether a BM is a CSBM:

- BM Day 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 for that day.
- Association with a Sense of Complete Evacuation
  This is assessed by the patient answering the following eDiary question for each BM:
  "Did you feel like you completely emptied your bowels after your bowel movement?"

  1=Yes
  2=No

- Day of Any Laxative Use
  Patients will report use of bisacodyl or other laxatives, suppositories, or enemas on an event-driven basis. An evening report will ask whether the patient entered all laxative use for that day, and collect bisacodyl or other laxatives, suppositories, or enemas not previously entered for that day.

Each day of the Pretreatment, Treatment, and RW Periods, the patient will complete eDiary entries on an event-driven basis to report BMs, and whether the BM was associated with a sense of complete evacuation. (The patient is also asked to provide assessments of stool consistency and straining, which are described below.) The patient will also complete eDiary entries on an event-driven basis to report use of protocol-permitted bisacodyl (tablets or suppositories) or other laxatives, suppositories, or enemas to treat their symptoms. Patients will complete a daily evening report to enter any BMs and laxative usage not previously reported by the patient for that day (recall is limited to 24 hours or to the time of the previous evening's report).

Spontaneous Bowel Movement

The SBM assessment is based on the eDiary questions (described above) that determine whether a BM is an SBM:

- BM day and time
- Day of Any Laxative Use

Stool Consistency (BSFS)

Patient assessment of stool consistency will be collected using two questions by eDiary entry on an event-driven basis (ie, for each BM reported in the eDiary).

For each BM, the patient assesses his/her stool using the BSFS (Appendix 4) 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.(14) The 7-point ordinal BSFS is provided below (see Appendix 4 for full scale including pictures):

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

1=Separate hard lumps like nuts (difficult to pass)
2=Like a sausage but lumpy
3=Like a sausage but with cracks on the surface
4=Like a sausage or snake, smooth and soft

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

6=Fluffy pieces with ragged edges, a mushy stool

7=Watery, no solid pieces (entirely liquid)

The patient also assesses his/her stool using a 5-point ordinal scale described below.

Straining

Patient assessment of straining will be collected by eDiary entry on an event-driven basis (ie, for each BM reported in the eDiary). For each BM, degree of straining will be assessed by the patient using the following 5-point ordinal scale:

"How much did you strain during your bowel movement?"

1=Not at all

2=A little (mild straining)

3=A fair amount (moderate straining)

4=A large amount (severe straining)

5=An extreme amount (extremely severe straining)

Stool Consistency (5-point ordinal scale)

In addition to the patient assessment of stool consistency using the BSFS (described above), patients will also assess stool consistency by daily eDiary entry on an event-driven basis 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

Weekly Patient Assessment of Constipation Severity

Patient assessment of constipation severity will be reported weekly by eDiary entry. The rating of constipation severity during the previous 7 days on a 5-point ordinal scale will be provided by the patient answering the following question:

"On average, how would you rate your constipation during the past 7 days?"

1=None
2=Mild
3=Moderate
4=Severe
5=Very severe

Weekly Patient Assessment of IBS Symptom Severity

Patient assessment of IBS symptom severity will be reported weekly by eDiary entry. The rating of IBS symptom severity during the previous 7 days on a 5-point ordinal scale will be provided by the patient answering the following question:

"On average, how would you rate your IBS symptoms during the past 7 days?"

1=None
2=Mild
3=Moderate
4=Severe
5=Very severe

Weekly Patient Assessment of Adequate Relief

Patient assessment of adequate relief of IBS symptoms 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 symptoms 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 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

8.1.4 Health Economics and Outcomes Research Assessments

IBS-QOL

The IBS-QOL is an instrument for assessing the impact of IBS on a patient's quality of life (Appendix 5).(15) The IBS-QOL 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.

CDC Healthy Days Core Module (CDC HRQOL-4)

The CDC Healthy Days Core Module (CDC HRQOL-4; Appendix 6) assesses a person's perceived health status through 4 questions that assess self-rated health, numbers of recent days when physical health or mental health was not good, and number of recent days with limitations due to poor physical or mental health.(16) For the CDC HRQOL-4 items, recent is defined as during the past 30 days. The CDC HRQOL-4 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.

8.1.5 Completion of eDiary Assessments

To ensure patient compliance with the completion of the evening eDiary report, alarms occurring at 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.

8.2 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.

8.2.1 Adverse Events

8.2.1.1 Definitions

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.

Serious Adverse Event

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 trial 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.

Treatment-emergent Adverse Event

A treatment-emergent AE (TEAE) is an event that emerges, or a preexisting event that worsens, any time after administration of the first dose of study drug (Day 1).

8.2.1.2 Classification of an Adverse Event

8.2.1.2.1 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 subject reported nausea lasting 3 days, one start date and stop date should be recorded along with the maximum intensity experienced for that event over that 3-day timeframe).

8.2.1.2.2 Relationship to Study Drug

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 Table 2.

Table 2. Adverse Event Causality

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 subject'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

8.2.1.2.3 Laboratory Abnormalities

The investigator will review clinical laboratory values for significance and consideration as an AE. All of the following laboratory abnormalities should be captured as AEs:

- Any laboratory test result that meets criteria for an SAE
- Any laboratory abnormality that results in discontinuation
- Any laboratory abnormality that requires the patient to receive specific corrective therapy
- Any laboratory abnormality that the investigator considers to be clinically significant

Ongoing abnormal laboratory values/conditions that are being treated at baseline will be captured as an AE if the condition increases in severity and/or frequency during the course of the trial or if the condition requires more frequent treatment. If a patient is treated for an abnormal laboratory value just before the Screening Visit, then the medical history should reflect the severity of the condition before treatment.

8.2.1.3 Procedures for Recording Adverse Events

The investigator will record all AEs from the time informed consent is obtained until completion of trial participation (eg, early termination or Week 16/EOT Visit). At each trial visit, the investigator will inquire about the occurrence of AEs/SAEs since the last visit.

The occurrence of an AE or SAE may come to the attention of trial center personnel during trial visits and interviews of a trial patient presenting for medical care, or upon review by a site monitor. All AEs including SAEs will be captured on the appropriate eCRF. Information to be collected includes event description, time of onset, clinician's assessment of severity, relationship to study drug (assessed only by those with the training and authority to make a diagnosis), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship. All AEs will be followed to adequate resolution or stabilization.

Any medical condition that is present at the time that the patient is screened and does not deteriorate (worsen in severity and/or frequency) should be recorded as Medical History and not as an AE. However, if the patient's condition deteriorates at any time during the trial, it will be recorded as an AE. AEs characterized as intermittent require documentation of onset and duration of each episode.

Pretreatment AEs will be collected and captured in the patient's source documentation from the time the patient signs the ICF until the patient receives study drug. Pretreatment AEs in randomized patients will additionally be entered on the AE page of the patient's eCRF.

Laboratory abnormalities, changes in vital signs, and physical examination findings should be considered AEs and reported on the AE page of the patient's eCRF only if the investigator considers them clinically significant and/or they necessitate intervention.

8.2.1.4 Procedures for Collecting and Reporting Serious Adverse Events

The investigator or designee is to report any SAE to Ironwood Pharmacovigilance (see below) using the SAE Form within 24 hours of becoming aware of the event that occurred during the reporting period.

Ironwood Pharmacovigilance Contact Information
Ironwood Pharmaceuticals, Inc.
clinicalsafety@ironwoodpharma.com
Fax: 1-617-933-7688

The SAE Form, which is to be completed in English and signed by the investigator (or designee), is to include as much information as possible, but at a minimum must contain the following:

- SAE term
- Serious criteria
- Severity
- Causality assessment
- Narrative explaining the context of the SAE outcome

If not all information on the SAE Form is available at the time of the initial report, follow-up SAE reports will be completed and submitted within the same reporting timelines as initial reports.

The investigator or designee is required to follow SAEs until resolution regardless of whether the patients are still participating in the trial. Resolution is defined as:

- Resolved with or without residual effects (sequelae)
- A return to baseline for a preexisting condition
- The investigator does not expect any further improvement or worsening of the event
- Fatal outcome: If an autopsy is performed on a deceased patient, the autopsy report and death certificate must be provided to Ironwood as soon as it is available.

8.2.1.4.1 Reporting of SAEs to the IRB

The investigator will receive prompt notification of SAEs, with the use of the study product, that are both unexpected and related, or any finding that suggests a significant risk for patients. The investigator will promptly inform the IRB of the notification and insert the notification in the Investigator's Regulatory Binder in accordance with local regulations.

All applicable expedited safety reports will be forwarded, by Ironwood, to the investigator.

The investigator will inform Ironwood of any local regulatory or IRB requirements not covered by the procedures in this or the prior section.

8.2.1.4.2 Reporting of Pregnancy

Any female patient who becomes pregnant while participating in the trial will be withdrawn from the trial.

Information on any pregnancies in female patients, or the female partner of a male patient, will be collected from the Screening Visit until the completion of the Week 16/EOT Visit.

If the patient or the female partner of a male patient becomes pregnant after receiving study drug during the course of trial, the investigator will collect and record the pregnancy information on the Pregnancy Reporting Form and submit it to Ironwood within 24 hours of learning of the pregnancy. (Note: If the female partner of a male patient becomes pregnant, the investigator must attempt to obtain consent to collect pregnancy information [including status of the newborn, if applicable] before reporting information to Ironwood). If not all information on the Pregnancy Reporting Form is available at the time of the initial report, follow-up Pregnancy Reporting Forms will be completed and submitted within 24 hours of becoming aware of new information.

The investigator is required to attempt follow-up on the pregnancy until the completion of the pregnancy. Information on the status of the mother and newborn will be forwarded to Ironwood within 24 hours of the investigator becoming aware. Generally, follow-up will be no longer than 6 to 8 weeks following the estimated delivery date.

While pregnancy itself is not considered to be an AE, any pregnancy complication or elective termination of a pregnancy for medical reasons will be an SAE and reported as such. A spontaneous abortion is always considered to be an SAE and will be reported as such. Furthermore, any SAE occurring as a result of a post-trial pregnancy and considered reasonably related to the investigational product by the investigator, will be reported to Ironwood as described in Section 8.2.1.4. While the investigator is not obligated to actively seek this information in former trial participants, he or she may learn of an SAE through spontaneous reporting.

8.2.2 Clinical Laboratory Determinations

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 trial 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). Clinical significance of a positive urine drug screen will be assessed by the investigator; positive results for cocaine, barbiturates, and cannabinoids are exclusionary.

- **Pregnancy test**: Serum human chorionic gonadotropin pregnancy test (for women of childbearing potential only) should be conducted at the Screening Visit and Week 16/EOT Visit. A negative urine pregnancy test must be documented at the Randomization Visit and the Week 12/ETP Visit for the patient 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 trial.

The investigator must review each laboratory report, document this review, and record any clinically relevant changes occurring during the trial 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 trial 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.

8.2.3 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. Blood pressure 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).

8.2.4 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.

A rectal examination should be performed during the Screening Period in all patients who do not require a colonoscopy (refer to Appendix 3). After the Screening Period, the rectal examination is optional and may be performed at the discretion of the investigator. For all physical examinations, the breast and genitourinary examinations are optional and may be performed at the discretion of the investigator.

8.2.5 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 questions to determine whether they also meet the Rome IV criteria for IBS, as 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

Rome IV diagnosis has no bearing on patient eligibility to participate in the trial (patients will be enrolled based on Rome III criteria for IBS, as detailed in Section 6.1).

8.2.6 Prior IBS-C Symptom Management Assessment

The prior IBS-C symptom management assessment (Appendix 7) will be performed at the Screening Visit, and includes prior treatments taken for IBS-C, lifestyle and diet modifications for alleviating the symptoms of IBS-C, assessment of satisfaction with prior interventions' ability to relieve bowel and abdominal symptoms, and primary reason for stopping use.

9. DATA QUALITY ASSURANCE

9.1 DATA MONITORING

Before any patient enters the trial, 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 trial 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 trial 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 will make available to the site monitor the computer that accesses the eCRFs during monitoring visits. 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.

9.2 DATA RECORDING AND DOCUMENTATION

All data collected in the context of this trial 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. Trial records (eg, essential documents [commonly called regulatory documents], correspondence) will be retained at the trial center, along with adequate source documentation, according to FDA and ICH requirements. All trial records must be available for inspection by Ironwood, its authorized designee, and the FDA.

Data collection will involve the use of the [redacted] EDC system, 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 trial 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.

10. STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

10.1 ANALYSIS POPULATIONS

10.1.1 Screened Population

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

10.1.2 Intent-to-Treat Population

The Intent-to-Treat (ITT) Population consists of all randomized patients.

10.1.3 Randomized Withdrawal (RW) Population

The RW Population consists of all patients who were rerandomized or allocated to study drug upon completion of the Treatment Period.

10.1.4 Safety Population

The Safety Population consists of all patients who received at least one dose of study drug.

10.2 GENERAL METHODS

Efficacy outcomes from the Treatment Period will be based on the ITT Population and will be summarized by treatment group assigned at the Randomization Visit. Efficacy outcomes from the RW Period only or from the Combined Treatment and RW Period will be based on the RW Population and will be summarized by treatment sequence determined at the Week 12/ETP Visit. Treatment periods will be summarized for safety in the same manner as efficacy; however, safety summaries (Safety Population) will be based on the actual treatment received.

Descriptive statistics including the number of patients, mean, minimum and maximum, and standard deviation (SD) will be calculated for continuous variables. Frequencies and percentages for each category will be calculated for categorical variables. Percentages will be based on the total number of non-missing values. The number missing will be presented, but without a percentage. All hypothesis tests will be two-sided with a 5% significance level, and 95% confidence intervals (CIs) will be used, unless stated otherwise.

10.2.1 Patient Disposition

Screen failures (ie, patients who entered the Screening Period but not the Pretreatment Period) and pretreatment failures (ie, patients who entered the Pretreatment Period but were not randomized) and reason for failure will be tabulated.

Patient counts for the ITT Population will be provided by trial center and geographic region.

Descriptive summaries will be presented for the ITT Population, the Safety Population, for those who completed the Treatment Period, and for those who prematurely discontinued (including the reason for premature discontinuation as recorded on the trial completion form).

Similar to the Treatment Period, descriptive summaries of patients who were rerandomized for the RW Period, completed the RW Period, and prematurely discontinued during the RW Period plus reasons for premature discontinuation will be presented for the RW Population.

10.2.2 Demographics and Baseline 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 ITT Populations. Baseline efficacy parameters (including SBM frequency, CSBM frequency, BSFS score, stool consistency, straining, abdominal score, abdominal bloating, abdominal discomfort, and abdominal pain) will be summarized descriptively for the ITT Population.

10.2.3 Protocol Deviations

Protocol deviations and Important Protocol Deviations (IPD) will be identified and documented for all ITT 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 trial even though they did not satisfy the entry criteria
- Those who developed withdrawal criteria during the trial but were not withdrawn
- Those who received the wrong treatment or incorrect dose
- Those who received an excluded concomitant treatment

10.2.4 Prior and Concomitant Medicines

Prior and concomitant medicines will be classified using the most current version of the WHO Drug Dictionary available at the start of the trial (or newer).

Prior medicines are defined as any medicines taken prior to the date of first dose of study drug. Concomitant medicines are defined as any medicines taken on or after the date of first dose of study drug during the defined period (Treatment/RW). Any medicines started after the date of last dose of study drug for the defined period (Treatment/RW) will be excluded from the summaries related to concomitant medicines for that period.

Prior medicine use, concomitant medicine in the Treatment Period, and concomitant medication use in the RW Period use will be summarized. Multiple medicines used by a patient in the same category (based on Anatomical-Therapeutic-Chemical classification) will be counted only once.

10.3 EXTENT OF EXPOSURE AND TREATMENT COMPLIANCE

10.3.1 Exposure to Study Drug

Exposure to study drug, calculated as the number of days from the date of first dose taken to the date of last dose taken, will be summarized by treatment group for the Treatment Period (Safety Population) and by treatment sequence for the RW Period (RW Population).

10.3.2 Measurement of Treatment Compliance

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

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. Similarly, descriptive statistics for the RW Period will be presented for the RW Population.

10.3.3 eDiary Compliance

eDiary compliance will be based on the percentage of complete eDiary entries made by a patient. A complete eDiary entry is defined as one in which the patient responds to every eDiary question asked in the evening report on that day. The questions that are asked weekly will not be included in the determination of a complete entry.

Descriptive statistics for eDiary compliance (%) and the patients with ≥80%/<80% complete eDiary entries during the Pretreatment, Treatment, and RW Periods will be presented.

10.4 EFFICACY ANALYSES

All efficacy analyses to address the objectives of the trial will be based on the ITT Population for outcomes occurring during the Treatment Period. Additional exploratory analyses will be defined and clearly specified for the RW Period.

Baseline values for efficacy 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 up to the time of randomization. Baseline values for patient symptom severity parameters (eg, abdominal symptoms [pain, bloating, discomfort], constipation severity) will be the average of the non-missing severity scores reported during this period. The baseline CSBM and SBM weekly rates will be derived based on the number of CSBMs and SBMs a patient had during this period. Baseline BSFS score, stool consistency, and straining will be calculated as the average of the non-missing values associated with the SBMs reported by the patient during this period.

Trial centers will be pooled together by geographic region (details to be provided in the Statistical Analysis Plan [SAP]). In lieu of trial center, geographic region will be used in analyses adjusting for center-to-center variability.

Sensitivity analyses of the key endpoints will include, but are not limited to, a multiple imputation approach. If the normality assumption of the errors is violated for the key mixed model with repeated measures (MMRM) or analysis of covariance (ANCOVA) analyses, ranked or other appropriate analyses will be applied.

The overall family-wise Type I error rate for the primary and secondary efficacy analyses will be controlled at a two-sided 0.05 significance level.

10.4.1 Primary Efficacy Endpoint

The primary efficacy endpoint is:

- **Change from Baseline in Abdominal Score at Each Week**
  A patient's daily Abdominal Score is calculated as the average of the daily patient assessments of abdominal bloating at its worst, abdominal discomfort at its worst, and abdominal pain at its worst. If 2 or more of the individual daily abdominal symptoms are missing, then the Abdominal Score for that day will be missing. The weekly Abdominal Score is the average of the non-missing Abdominal Scores during each week (Weeks 1-12) in the trial. The baseline Abdominal Score is the average of the non-missing Abdominal Scores during the 14-day Pretreatment Period and the day of the Randomization Visit reported prior to randomization. Change from baseline will be calculated for each week as the weekly score minus the baseline score.

10.4.2 Primary Efficacy Analysis

Inferential testing between the linaclotide 290 µg and placebo groups with regard to the change from baseline in abdominal score over the Treatment Period will be evaluated employing a mixed model with repeated measures (MMRM) framework with week (categorical), treatment, geographic region, and week-by-treatment fixed effects, patient as the random effect, and baseline value as a covariate. An unstructured covariance structure will be utilized. Descriptive statistics based on the MMRM model will include least-squares (LS) mean change from baseline for each treatment, the LS mean difference between linaclotide 290 µg and placebo, corresponding 95% CIs, and the p-value associated with the comparison.

10.4.3 Secondary Efficacy Endpoints

The secondary efficacy endpoints are:

- Change from Baseline in 12-week Abdominal Score (Cumulative Distribution Function [CDF])

  Daily Abdominal Score is calculated as described for the primary endpoint in Section 10.4.1. The 12-week Abdominal Score is the average on the non-missing Abdominal Scores reported over the course of the Treatment Period. The baseline Abdominal Score is the average of the non-missing Abdominal Scores during the 14-day Pretreatment Period and the day of the Randomization Visit reported prior to randomization. Change from baseline will be calculated as the 12-week score minus the baseline score.

- 6/12 Week Abdominal Score Responder

  A 6/12 Week Abdominal Score Responder is a patient who meets the Weekly Abdominal Score Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly Abdominal Score Responder is a patient who has an improvement from baseline of at least 2 points (ie, a -2 change from baseline) in the respective weekly abdominal score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly Abdominal Score Responder for that week.

10.4.4 Secondary Efficacy Analysis

The CDFs for linaclotide 290 µg and placebo will be estimated for the Change from Baseline in 12-week Abdominal Score endpoint. The CDFs will be compared using the Wilcoxon rank sum test with Hodges-Lehmann estimator for the median difference.

A secondary time-course analysis of the Change from Baseline in Abdominal Score (see primary endpoint definition in Section 10.4.1) will also be conducted. Using the MMRM framework defined in the primary analyses, treatment difference between linaclotide 290 µg and placebo will be assessed at each individual week. For each week, descriptive statistics based on the MMRM model will include the LS mean change from baseline for each treatment, the LS mean difference between linaclotide 290 µg and placebo, corresponding 95% CIs, and the p-value associated with the treatment comparison.

For the secondary responder endpoint, the proportion of responders in the linaclotide 290 µg group will be compared to the proportion of responders in the placebo group using a Cochran-Mantel-Haenszel (CMH) test controlling for geographic region. The number and percent of responders, the difference in responder rates between the linaclotide 290 µg and placebo groups, the odds ratio relative to placebo, all corresponding 95% CIs, and the p-value associated with the CMH test will be presented.

10.4.5 Controlling for Multiplicity

The overall family-wise Type I error rate for the primary and secondary efficacy analyses will be controlled at the α=0.05 level by employing a fixed-sequence procedure as described below. Following the fixed-sequence procedure, if the primary hypothesis between the placebo group and the linaclotide 290 µg group is statistically significant (α=0.05) in the ITT Population, then the primary objective of the trial will have been achieved and the next hypothesis can be tested; otherwise, testing will stop. The testing of each sequential hypothesis is conditional on all the previous hypotheses being rejected at the 0.05 level of significance. If a hypothesis is not rejected at the 0.05 level of significance, then all remaining hypotheses are deemed not statistically significant. All hypothesis testing will be two-sided.

1. Linaclotide vs. placebo – Change from Baseline in Abdominal Score (primary efficacy analysis)
2. Linaclotide vs. placebo – Change from Baseline in 12-week Abdominal Score (CDF) (secondary efficacy analysis)
3. Linaclotide vs. placebo – 6/12 Week Abdominal Score Responder (secondary efficacy analysis)
4. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 12
5. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 10
6. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 8
7. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 6
8. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 4
9. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 2
10. Linaclotide vs. placebo – Change from Baseline in Abdominal Score at Week 1

10.4.6 Additional Efficacy Endpoints

Additional efficacy endpoints will be explored outside of the formal testing procedures described in Section 10.4.5. These endpoints will include:

- 6/12 Week Abdominal Pain and Constipation (APC) +1 Responder

  A 6/12 Week APC +1 Responder is a patient who meets the Weekly APC +1 Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly APC +1 Responder is a patient who has an increase from baseline of at least 1 in the respective CSBM weekly rate and has a decrease from baseline of at least 30% in the respective weekly abdominal pain score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a responder for that week.

- Change from Baseline in Abdominal Pain at its Worst at Each Week

  Abdominal pain is measured daily using an 11-point NRS. For each week on treatment, the weekly abdominal pain score is the mean of the non-missing daily values.

- Change from Baseline in CSBM Frequency Rate at Each Week

  A patient's weekly CSBM frequency rate is the CSBM rate (CSBMs/week) calculated for that week.

- Change from Baseline in SBM Frequency Rate at Each Week

  A patient's weekly SBM frequency rate is the SBM rate (SBMs/week) calculated for that week.

- Change from Baseline in BSFS (Stool Consistency) at Each Week

  Stool consistency is measured using the 7-point BSFS. The patient's BSFS score for each week on treatment is the mean of the non-missing BSFS scores from the SBMs reported by the patient during the week.

- Change from Baseline in Stool Consistency at Each Week

  Stool consistency is measured using a 5-point ordinal scale. The patient's stool consistency score for each week on treatment is the mean of the non-missing stool consistency scores from the SBMs reported by the patient during the week.

- Change from Baseline in Straining at Each Week

  Straining is measured using a 5-point ordinal scale. The patient's straining score for each week on treatment is the mean of the non-missing straining scores from the SBMs reported by the patient during the week.

- Change from Baseline in Abdominal Discomfort at its Worst at Each Week

  Abdominal discomfort is measured daily using an 11-point NRS. For each week on treatment, the weekly abdominal discomfort score is the mean of the non-missing daily values.

- Change from Baseline in Abdominal Bloating at its Worst at Each Week

  Abdominal bloating is measured daily using an 11-point NRS. For each week on treatment, the weekly abdominal bloating score is the mean of the non-missing daily values.

- Percent Change from Baseline in Abdominal Pain at its Worst at Each Week

  Percent change will be calculated as 100 * the Change from Baseline in Abdominal Pain at Worst at each week divided by the baseline abdominal pain score.

- Change from Baseline in Percent of Days with Use of Rescue Medicine

  The percent of days using per-protocol rescue medicine or any other laxative, suppository, or enema during at each week be calculated as 100 * the number of days rescue medicine was used during the week divided by the total number of days on treatment.

- SBM Within 24 Hours After First Dose Responder

  A patient meets the SBM Within 24 Hours After First Dose Responder criteria if the amount of time between the time of first dose of study medication reported at the randomization visit and the time of first reported SBM is less than or equal to 24 hours.

- 6/12 Week Abdominal Pain 30% Responder

  A 6/12 Week Abdominal Pain 30% Responder 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. For each week in the Treatment Period, a Weekly Abdominal Pain 30% Responder is a patient who has a decrease from baseline of at least 30% in the respective weekly abdominal pain score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly Abdominal Pain 30% Responder for that week.

- 6/12 Week Abdominal Pain Responder

  A 6/12 Week Abdominal Pain Responder is a patient who meets the Weekly Abdominal Pain Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly Abdominal Pain Responder is a patient who has an improvement from baseline of at least 2 points (ie, a -2 change from baseline) in the respective weekly abdominal pain score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly Abdominal Pain Responder for that week.

- 6/12 Week Abdominal Discomfort Responder

  A 6/12 Week Abdominal Discomfort Responder is a patient who meets the Weekly Abdominal Discomfort Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly Abdominal Discomfort Responder is a patient who has an improvement from baseline of at least 2 points (ie, a -2 change from baseline) in the respective weekly abdominal discomfort score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly Abdominal Discomfort Responder for that week.

- 6/12 Week Abdominal Bloating Responder

  A 6/12 Week Abdominal Bloating Responder is a patient who meets the Weekly Abdominal Bloating Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly Abdominal Bloating Responder is a patient who has an improvement from baseline of at least 2 points (ie, a -2 change from baseline) in the respective weekly abdominal bloating score. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly Abdominal Bloating Responder for that week.

- 6/12 Week CSBM Responder

  A 6/12 Week CSBM Responder is a patient who meets the Weekly CSBM Responder criteria for at least 6 out of the 12 weeks of the Treatment Period. For each week in the Treatment Period, a Weekly CSBM Responder is a patient who has an increase from baseline of at least 1 in the respective CSBM weekly rate. If a patient did not enter information into the eDiary on at least 4 days for a particular Treatment Period week, the patient will not be considered a Weekly CSBM Responder for that week.

- Treatment Satisfaction

  Treatment satisfaction is measured on a 5-point ordinal scale. Treatment satisfaction will be analyzed separately for each Treatment Period week.

- Adequate Relief

  Assessment of adequate relief during the previous 7 days is measured on a binary scale (yes/no). Assessment of adequate relief will be analyzed separately for each Treatment Period week.

- 6/12 Week Adequate Relief Responder

  A 6/12 Week Adequate Relief Responder is a patient who reports Adequate Relief for at least 6 out of the 12 weeks of the Treatment Period.

- Change from Baseline in Constipation Severity

  Constipation severity is measured weekly using a 5-point ordinal scale. Change from baseline will be calculated at each week.

- Change from Baseline in IBS Symptom Severity

  IBS symptom severity is measured weekly on a 5-point ordinal scale. Change from baseline will be calculated at each week.

10.4.7 Additional Efficacy Analysis

Change-from-baseline endpoints and Treatment Satisfaction will be analyzed utilizing the same MMRM methods as described for the primary analysis in Section 10.4.2, and by visit as described in Section 10.4.4. Responder endpoints will be analyzed using the methodology defined in Section 10.4.4.

10.4.8 Randomized Withdrawal Period Endpoints

For the RW Period, statistical analyses will be presented by treatment; there are 3 possible treatment sequences:

1. 290 µg-290 µg (linaclotide 290 µg administered in the Treatment Period, followed by linaclotide 290 µg in the RW Period)
2. 290 µg-Placebo (linaclotide 290 µg administered in the Treatment Period, followed by placebo in the RW Period)
3. Placebo-290 µg (placebo administered in the Treatment Period, followed by linaclotide 290 µg in the RW Period)

The following endpoints will be calculated for the RW Period following the methods described for the primary and secondary endpoints in Sections 10.4.2 and 10.4.4:

- Change from Baseline in Abdominal Score at Each Week
- Change from Baseline in Abdominal Bloating at its Worst at Each Week
- Change from Baseline in Abdominal Discomfort at its Worst at Each Week
- Change from Baseline in Abdominal Pain at its Worst at Each Week
- Change from Baseline in CSBM Frequency Rate at Each Week
- Change from Baseline in SBM Frequency Rate at Each Week
- Change from Baseline in BSFS (Stool Consistency) at Each Week
- Change from Baseline in Stool Consistency at Each Week
- Change from Baseline in Percent of Days with Use of Rescue Medicine

10.4.9 Randomized Withdrawal Period Analyses

All RW Period endpoints will be summarized for the RW Period only and for the Combined Treatment and RW Period using descriptive statistics and 95% CIs for each treatment sequence (sequences are defined in Section 10.4.8). No statistical inference will be performed. Baseline for all RW Period endpoints will be defined as described in Section 10.4.

10.5 HEALTH ECONOMICS AND OUTCOMES RESEARCH ANALYSES

Health economics and outcomes research analyses will be based on the ITT Population for the Treatment Period and on the RW Population for the RW Period.

10.5.1 IBS-QOL

The IBS-QOL parameters consist of the overall average score and eight subscale scores (ie, dysphoria, interference with activity, body image, health worry, food avoidance, social reaction, sexual, and relationships). For the Week 12/ETP Visit and the Week 16/EOT Visit, change from baseline in the IBS-QOL overall average score and the subscale scores will be analyzed using an ANCOVA model with fixed effect terms for treatment group and geographic region and the corresponding baseline IBS-QOL score as a covariate.

10.5.2 CDC HRQOL-4

The CDC HRQOL-4 (the Healthy Days Core Module) consists of 4 questions. Self-rated general health, physically unhealthy days, mentally unhealthy days, and poor physical/mental days are collected. Unhealthy days, defined as the minimum of the sum of physically unhealthy days and mentally unhealthy days, or 30 days, will be calculated and reported. Descriptive summaries for the derived unhealthy days and the change from baseline will be summarized descriptively at each visit. Treatment difference between the change from baseline score will be performed using ANCOVA model with fixed effect terms for treatment group and geographic region and the corresponding baseline measure as a covariate.

10.6 SAFETY ANALYSES

All safety parameters will be analyzed descriptively in accordance with the general methods described in Section 10.2.

10.6.1 Adverse Events

Adverse Event Verbatim Terms will be coded in the EDC system against the most current version of MedDRA available at the start of the trial (or newer).

An AE that occurs during the Treatment Period will be considered a treatment-emergent AE (TEAE) for the Treatment Period if it was not present prior to the date of the first dose of double-

blind study drug, or was present prior to the date of the first dose but increased in severity during the Treatment Period.

Similarly, an AE that occurs on or after the date of the first dose of double-blind study drug for the RW Period (up to 1 day after the last dose of double-blind study drug) will be considered a TEAE for the RW Period if it was not present prior to the date of first dose of study drug for the Treatment Period, or was present prior to the to the date of the first dose for the Treatment Period but increased in severity on or after the date of the first dose for the RW Period.

An AE that occurs on or after the date of the first day of dosing in the RW Period (up to 1 day after the last dose of double-blind study drug) will be considered a newly-emergent adverse event (NEAE) for the RW Period if it was not present prior to the date of the first calendar day of dosing in the RW Period, or was present prior to the date of the first calendar day of dosing in the RW Period, but increased in severity on or after the date of the first calendar day of dosing in the RW Period.

If more than one AE is reported prior to the first dose of double-blind study drug and coded to the same preferred term, the AE with the greatest severity will be used as the benchmark for comparison with the AEs occurring during the Treatment and RW Periods that were also coded to that preferred term. An AE that occurs more than 1 day after the last dose of double-blind study drug will not be counted as a TEAE or NEAE.

For the Treatment and RW Periods, the number and percentage of patients reporting TEAEs in each treatment group and treatment sequence, respectively, will be tabulated by system organ class (SOC) and preferred term; by SOC, preferred term, and severity; and by SOC, preferred term, and relationship to study drug. If a patient has more than one TEAE coded to the same preferred term, the patient will be counted only once for that preferred term by identifying those TEAEs with the highest severity and the closest relationship to study drug. Similar tabulations will be presented for the number and percentage of patients reporting NEAEs in the RW Period by treatment sequence.

The distribution of TEAEs by severity and relationship to study drug will be summarized by treatment group for the Treatment Period. Similarly, the distribution of TEAEs and NEAEs by severity and relationship to study drug will be summarized by treatment sequence for the RW Period.

The incidence of common TEAEs, on-therapy SAEs, and AEs leading to premature discontinuation of study drug will be summarized by preferred term and treatment group for the Treatment Period and treatment sequence for the RW Period. In addition, the incidence of fatal SAEs (ie, events that caused death), if any, will be summarized separately by treatment group for the Treatment Period and treatment sequence for the RW Period, and preferred term.

Listings will be presented for patients with SAEs, patients with AEs leading to discontinuation, and patients who die (if any).

10.6.2 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 the Treatment Period and by treatment sequence for the Combined Treatment and RW Period, 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 Treatment Period and by treatment sequence for the RW Period and the Combined Treatment and RW Period. The criteria for PCS laboratory values will be detailed in the SAP. 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 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, trial center, and baseline and post-baseline values. A listing of all AEs for patients with PCS laboratory values will also be provided.

10.6.3 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/trial will be presented by treatment group for the Treatment Period and by treatment sequence for the Combined Treatment and RW Period.

The number and percentage of patients with PCS post-baseline vital signs will be tabulated by treatment group for the Treatment Period and by treatment sequence for the Combined Treatment and RW 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 will be detailed in the SAP. The percentages will be calculated relative to the number of patients with baseline values and at least 1 assessment in the corresponding post-baseline 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, trial center, and baseline and post-baseline values. A listing of all AEs for patients with PCS vital sign values will also be provided.

10.7 INTERIM ANALYSIS

No interim analysis is planned for this trial.

10.8 DETERMINATION OF SAMPLE SIZE

The sample size of 600 patients (300 patients per treatment group) was chosen to ensure adequate power for testing the fixed-sequence procedure for the primary and secondary efficacy endpoints as defined in Section 10.4.5.

The power calculations for the primary endpoint are based on the placebo and linaclotide 290 µg treatment groups from the Phase 3 trial LIN-MD-31. The patients in LIN-MD-31 are considered representative of the patient population for this trial. Using a resampling with replacement-based simulation (1000 iterations) and controlling for multiplicity as outlined above, the trial will have >99% power to reject the primary endpoint and ~94% power to reject all primary and secondary hypotheses defined in the testing process.

11. CHANGES IN THE CONDUCT OF THE TRIAL OR PLANNED ANALYSES

Any amendment to this protocol will be provided to the investigator in writing by Ironwood or designee. No protocol amendment regarding reportable deviations as defined by the IRB may be implemented (with the exceptions noted below) before it has been approved by the IRB, submitted by the Sponsor to the FDA, and the signature page, signed by the investigator, has been received by Ironwood or designee. If the protocol is amended to eliminate or reduce the risk to patients, the amendment may be implemented before IRB review and approval. However, the IRB must be informed in writing of such an amendment, and approval must be obtained within reasonable time limits. Deviating from the protocol is permitted only if absolutely necessary for the safety of the patients and must immediately be reported to Ironwood or designee.

12. TRIAL SPONSORSHIP

12.1 TRIAL TERMINATION

Ironwood reserves the right to terminate the trial in its entirety or at a specific center at any time.

12.2 REPORTING AND PUBLICATION

All data generated in this trial will be the property of Ironwood and its partner, Allergan, Plc. An integrated clinical and statistical report will be prepared at the completion of the trial.

Publication of the results by the investigator will be subject to mutual agreement between the investigator and Ironwood.

13. INVESTIGATOR OBLIGATIONS

13.1 DOCUMENTATION

The investigator must provide the following to Ironwood or designee prior to the start of the trial in accordance with ICH E6 and FDA regulations:

- A completed and signed Form FDA 1572. If, during the course of the trial, any changes are made that are not reflected on Form FDA 1572, a new Form FDA 1572 must be completed and returned to Ironwood or designee for submission to the FDA
- A fully executed Clinical Trial Agreement
- The curricula vitae for the investigator and all sub-investigators listed on Form FDA 1572, including a copy of each physician's license
- A copy of the initial IRB approval for conducting the trial. If the trial is ongoing, renewals must be submitted at yearly intervals. All amendments to the protocol must be submitted and approved by the IRB, as stated in Section 1.1
- A copy of the IRB-approved ICF
- A copy of the IRB-approved HIPAA authorization form
- A list of the IRB members **or** the DHHS general assurance number
- A copy of the laboratory reference ranges
- The Investigator's Statement page in this protocol signed and dated by the investigator
- Financial disclosure agreements completed and signed by the investigator and each sub-investigator listed on Form FDA 1572. If there are any relevant changes, the investigator (and any sub-investigator) will provide an updated financial disclosure agreement to the Sponsor at the time of the change, and up to 1 year after the completion of the trial

13.2 PERFORMANCE

The investigator must demonstrate reasonable efforts to obtain qualified patients for the trial.

13.3 USE OF INVESTIGATIONAL MATERIALS

The investigator will acknowledge that the drug supplies are investigational and as such must be used strictly in accordance with the protocol and only under the supervision of the investigator or sub-investigators listed on Form FDA 1572. Study drug must be stored in a safe and secure place. The investigator must maintain adequate records documenting the receipt, registration within IWRS, and disposition of all trial supplies. Ironwood or designee will supply forms on which to record the date the study drug was received and a dispensing record in which to record each patient's use. All unused study drug must be returned to Ironwood's designee. It is the investigator's responsibility to ensure that patients return their study drug.

13.4 CASE REPORT FORMS

All data relating to the study will be recorded on eCRFs to be provided by Ironwood or designee via the EDC system, or if applicable, paper CRFs. The eCRFs and paper CRFs are to be completed at the time of the patient's visit, except for results of tests performed outside the investigator's office. The investigator is responsible for verifying that all data entries on the eCRFs and paper CRFs are accurate and correct. The investigator must sign the completed eCRF before its submission to Ironwood or designee.

13.5 RETENTION AND REVIEW OF RECORDS

The investigator must maintain the documentation relating to this trial. If Ironwood or the FDA wishes to review any documentation relating to the trial, the investigator must permit access to such records.

Federal regulations require that the investigator retain a copy of all records that support eCRFs and paper CRFs for this trial (eg, ICFs, clinical laboratory reports, source documents, study drug dispensing records) for whichever of the following is the shortest:

- Two years following the date of approval by the FDA of the study drug for the purposes that were the subject of the investigation; or
- Five years following the date on which the results of the investigation were submitted to the FDA in support of, or as part of, a New Drug Application (NDA) for the study drug for the purposes that were the subject of the investigation

If the investigation does not result in the submission of the data in support of, or as part of, an NDA, records must be retained for two years following notification by Ironwood that the entire clinical investigation (not merely the investigator's portion) is completed, terminated, or discontinued or for two years following withdrawal of the Investigational New Drug Application or NDA.

If the investigator retires, relocates, or for other reasons withdraws from the responsibility of keeping the trial records, custody must be transferred to a person who will accept the responsibility. Ironwood must be notified in writing of the name and address of the new custodian.

13.6 PATIENT CONFIDENTIALITY

All patient records will only be identified by initials and PID number. Patients' names are not to be transmitted to Ironwood or its authorized designee. The investigator will keep a Master Patient List on which the PID number and the full name, address, and telephone number of each patient is listed.

14. APPENDICES

APPENDIX 1 ELEMENTS OF INFORMED CONSENT

Procedures will comply with 21 CFR, Parts 50 and 312. Signed informed consent will be obtained from each patient participating in a clinical research. This consent must include the following items:

- A statement that the trial involves research and an explanation of the purposes of the research; a description of the procedures to be followed and the identification of any procedures that are experimental; and the expected duration of the patient's participation
- A description of any reasonably foreseeable risks or discomforts to the patient
- A description of any benefits to the patient or to others that may reasonably be expected from the research. If the patient is to be paid for participating in the trial, the consent form must state the amount that he/she will receive and the schedule of payment (to ensure neither coercion nor undue influence)
- A disclosure of appropriate alternative procedures or courses of treatment, if any, that might be advantageous to the patient
- A statement describing the extent, if any, to which confidentiality of records identifying the patient will be maintained and noting the possibility that the FDA, Ironwood, the IRB, or an authorized CRO may inspect the records
- For research involving more than minimal risk, an explanation of whether any medical treatment is available if injury occurs and, if so, what it consists of or where further information may be obtained
- An explanation of whom to contact, including the relevant telephone number, for answers to pertinent questions about the research and the research patient's rights and whom to contact in the event of a research-related injury to the patient. (Note: in some cases, it may be necessary to identify a person other than the investigator as the contact. The guidance of the IRB may be required.)
- A statement that participation is voluntary, that refusal to participate will involve no penalty or loss of benefits to which the patient is otherwise entitled, and that the patient may discontinue participation at any time without penalty or loss of benefits to which the patient is otherwise entitled
- A statement that the particular treatment or procedures may involve risks to the patient (or to the embryo or fetus if the patient is, or may become, pregnant) that are at present unforeseeable

- The expected circumstances for which the patient's participation may be terminated by the investigator without regard to the patient's consent
- Any additional costs to the patient that may result from participation in the research
- The consequences of a patient's decision to withdraw from the research and procedures for an orderly termination of the patient's participation
- A statement that significant new findings developed during the course of the research that may relate to the patient's willingness to continue participation will be provided to the patient
- The approximate number of patients involved in the trial
- A statement of consent (eg, "I agree to participate...")
- A place for the patient's signature and date of signing
- A statement that a description of this clinical trial will be available on http://www.ClinicalTrials.gov

A copy of the signed consent form will be given to the patient.

APPENDIX 2 CONCOMITANT AND PROHIBITED MEDICATIONS

Rescue Medicine to Manage Constipation

Rescue Medicine, which will be selected by and dispensed to patients, will be a choice of 5-mg bisacodyl tablets or 10-mg bisacodyl suppositories. During the Pretreatment, Treatment, and RW Periods, patients may use dispensed, protocol-permitted laxatives (bisacodyl tablets or suppositories) as Rescue Medicine when at least 72 hours have passed since their previous BM or when their symptoms become intolerable. In order to qualify for randomization into the Treatment Period, patients must have refrained from using Rescue Medicine on the day before the Randomization Visit and on the day of the Randomization Visit prior to randomization. Patients must agree to refrain from using Rescue Medicine from the time they arrive at the clinic for the Randomization Visit through the day after randomization.

Prohibited Medicine

All medicine listed in the sections below ("1-day Washout" and "14-day Washout") are excluded during the Pretreatment, Treatment, and RW Periods. A 1-day washout means that the particular medicine is not allowed during the day before the Pretreatment Visit; a 14-day washout means that the particular medicine is not allowed during the 14 days before the Pretreatment Visit.

1-DAY WASHOUT ***(no medicine during the day before the Pretreatment Visit)***

1. Any over-the-counter or prescription laxative, suppository, or enema (eg, polyethylene glycol, lactulose, Fleet's) and any herbal or natural agent that a person might take for constipation. Note: The use of fiber, bulk laxatives, stool softeners (surfactants such as docusate), and probiotics is acceptable, provided the patient has been on a stable dose during the 30 days before the Screening Visit and plans to continue stable dosing throughout the trial.
2. Any medicine used to treat diarrhea (eg, bismuth subsalicylate, kaolin).
3. NSAIDs if taken for abdominal pain or discomfort.

14-DAY WASHOUT (*no medicine during the 14 days before the Pretreatment Visit)*

1. Drugs with known pharmacological activity at 5-hydroxytryptophan (HT)4, 5-HT2b or 5-HT3 receptors (eg, cisapride, tegaserod, ondansetron, tropisetron, granisetron, dolasetron, and mirtazapine).
2. Any treatment specifically taken for IBS-C or CIC alone or in combination, including lubiprostone (an approved chloride channel activator that enhances intestinal fluid secretion), linaclotide, plecanatide, colchicine, and misoprostol. Note: Patients may not have taken commercially available linaclotide or plecanatide, or participated in a linaclotide or plecanatide clinical study, during the 30 days before the Screening Visit.
3. Prokinetic agents (eg, metoclopramide, itopride, prucalopride, and domperidone).
4. Anti-cholinergic agents (eg, dicyclomine, flavoxate, scopolamine, hyoscyamine, propantheline, oxybutynin, tolterodine, solefenacin, darifenacin, and trospium). Note: inhaled ipratropium and tiotropium are permitted.
5. Bile acid sequestrants (eg, cholestyramine and colestipol).
6. Cholinomimetic agents (eg, bethanechol, pyridostigmine, tacrine, and physostigmine). Note: intraocular cholinomimetic agents (eg, pilocarpine) are permitted.
7. Antipsychotic agents (eg, risperidone, haloperidol, droperidol, chlorpromazine, perphenazine, all phenothiazines, quetiapine, olanzapine, and clozapine) unless the patient has been on a stable dose for 30 days before the Screening Visit and there is no plan to change the dose after the Screening Visit. Note: paliperidone is permitted without restriction.
8. Antidepressants unless the patient has been on a stable dose for 30 days before the Screening Visit and there is no plan to change the dose after the Screening Visit. Specifically included are the following:
    - Tricyclic antidepressants (eg, amitriptyline, imipramine, and nortriptyline);
    - Monoamine oxidase inhibitors (eg, furazolidone, isocarboxazid, pargyline, phenelzine, and selegiline tranylcypromine);
    - Selective serotonin reuptake inhibitors (eg, fluoxetine, sertraline, paroxetine, and citalopram);
    - Serotonin and norepinephrine reuptake inhibitors (eg, venlafaxine and desvenlafaxine succinate)
    - Others (eg, trazodone, and bupropion).

9. Calcium channel blocker verapamil unless the patient has been on a stable dose for 30 days before the Screening Visit and there is no plan to change the dose after the Screening Visit. Note: all other calcium channel blockers (eg, nifedipine, diltiazem, amlodipine, felodipine, nicardipine, nimodipine, nisoldipine) are permitted and may be used without restriction.
10. Oral and parenteral antibiotics (however, 1 standard regimen [up to 10 days] of oral antibiotics is permitted during the Treatment or RW Periods).
11. Any investigational or imported drugs that have not been approved for human use by the US FDA during the 30 days before the Screening Visit.
12. All narcotics either alone or in combination (eg, tramadol, codeine, morphine, propoxyphene, loperamide, diphenoxylate, and paregoric). Note: narcotics used as anesthesia for a colonoscopy require a 5-day wash-out prior to the patient entering into the Pretreatment Period.
13. Any medicine taken for the purpose of losing weight (eg, orlistat, phentermine, phendimetrazine, diethylpropion, benzphetamine, and sibutramine).
14. Any medicine that is known to cause diarrhea (eg, acarbose).
15. Proton pump inhibitors (eg, omeprazole, lansoprazole, esomeprazole, pantoprazole, rabeprazole) unless the patient has been on a stable dose for 30 days before the Screening Visit and there is no plan to change the dose after the Screening Visit.
16. Iron (as either a supplement or to treat iron-deficiency anemia) is acceptable, provided the patient has been on a stable dose for 30 days before the Screening Visit and plans to continue stable dosing throughout the trial.
17. Others: barbiturates (eg, butalbital and phenobarbital) and chronic oral or parenteral glucocorticoids (which must be discontinued at least 3 months before the Screening Visit; however, one 10-day course of oral or one injection of parenteral glucocorticoids is permitted during the Pretreatment, Treatment, or RW Periods). Pregabalin is acceptable, provided the patient has been on a stable dose during the 30 days before the Screening Visit and plans to continue stable dosing throughout the trial.

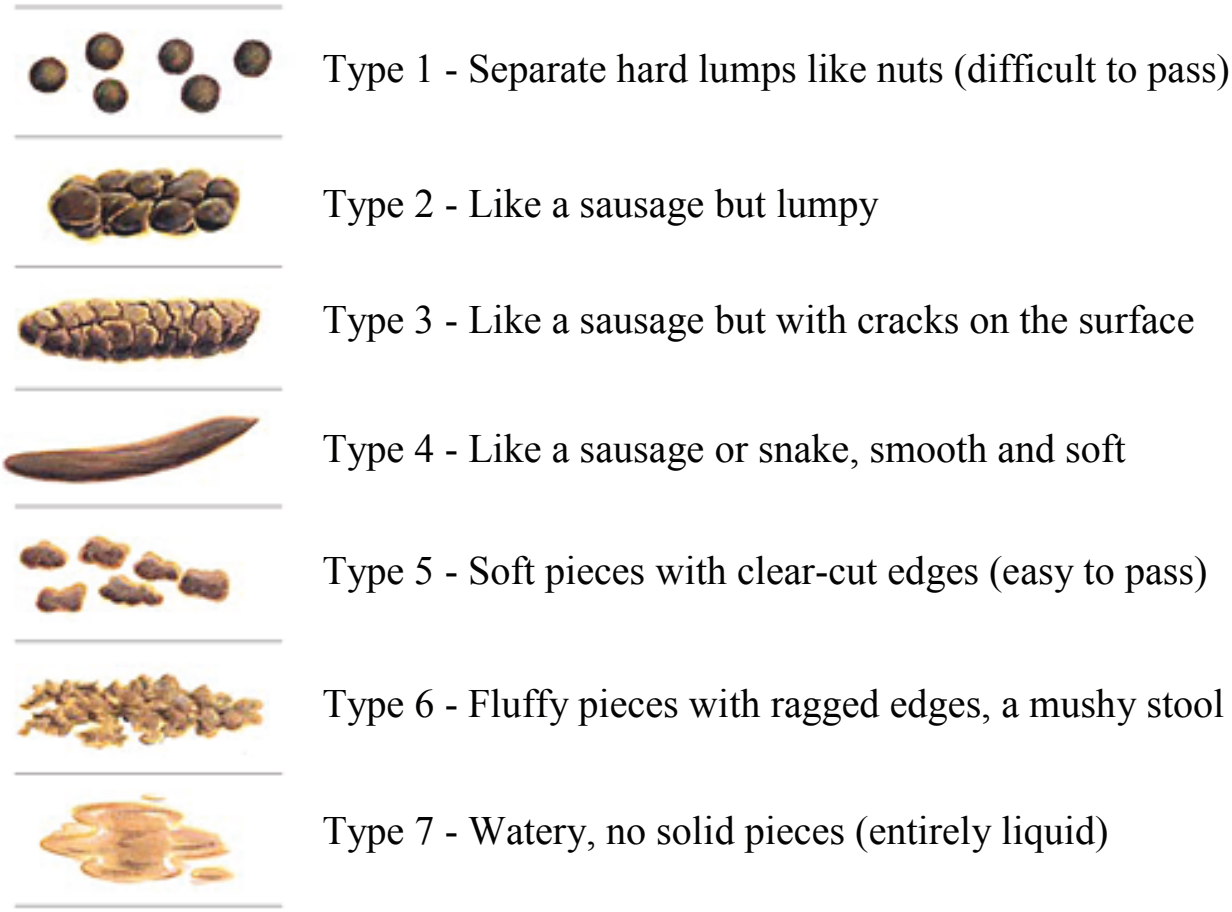
APPENDIX 3 SUMMARY OF AMERICAN GASTROENTEROLOGICAL ASSOCIATION GUIDELINES

1. Patients aged 50 years and older must have had a colonoscopy with negative findings during the 10 years before the Screening Visit. If there were polyps on the most recent colonoscopy, the patient may be enrolled, provided there were 2 or fewer small (<1 cm) tubular adenomas without appreciable villous tissue or high-grade dysplasia and provided the colonoscopy was performed during the 5 years before the Screening Visit. (Note: Patients who have only hyperplastic polyps are eligible.)
2. Patients who have a first-degree relative with colorectal cancer or adenomatous polyps diagnosed before age 60 or 2 first-degree relatives with colorectal cancer diagnosed at any age must have had a colonoscopy with negative findings during the 5 years before the Screening Visit. This applies to patients who are ≥40 years old and to patients <40 years old who are ≤10 years from the age when their youngest relative was found to have one of the conditions described above.
3. Patients who have a first-degree relative with colorectal cancer or adenomatous polyps diagnosed at age 60 or older or 2 second-degree relatives with colorectal cancer diagnosed at any age must have had a colonoscopy with negative findings during the 10 years before the Screening Visit. This applies to patients who are ≥40 years old and to patients <40 years old who are ≤10 years from the age when their youngest relative was found to have one of the conditions described above.
4. Patients of any age who have alarm symptoms must have had a colonoscopy with negative findings after the onset of the alarm symptoms and during the 5 years before the Screening Visit. Alarm symptoms include lower GI bleeding (rectal bleeding or heme-positive stool), iron-deficiency anemia, unexplained clinically-significant weight loss, and systemic signs of infection or colitis. In addition, patients with a family history of celiac disease or inflammatory bowel disease must have had a colonoscopy with negative findings during the five years before the Screening Visit.

Note: All information required by the Inclusion Criterion will be captured on the eCRFs. Patients who receive narcotics as anesthesia for a colonoscopy are eligible to enter the Pretreatment Period on the fifth day after the colonoscopy.

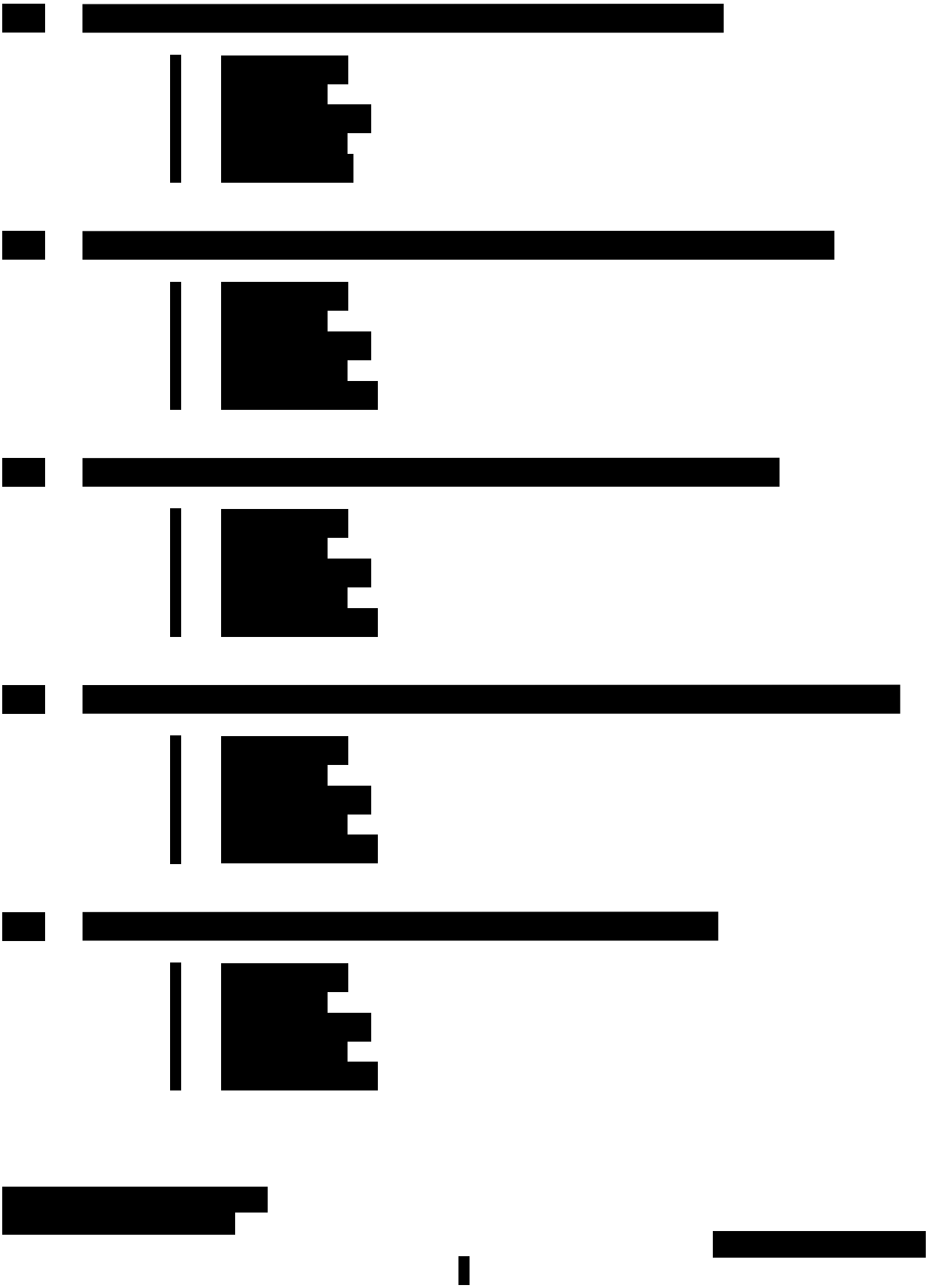
Source: Winawer, et al (2003)

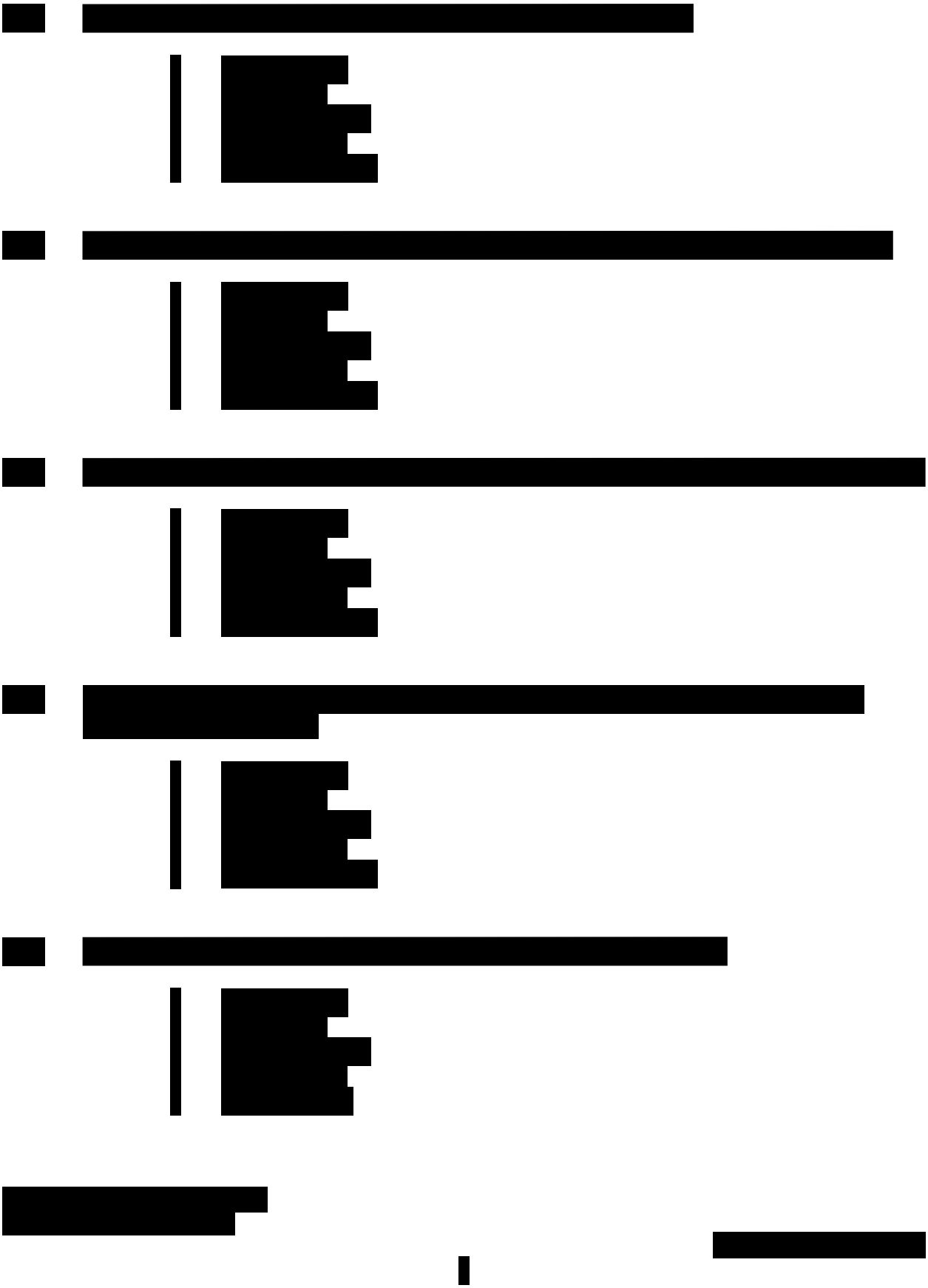
APPENDIX 4 BRISTOL STOOL FORM SCALE

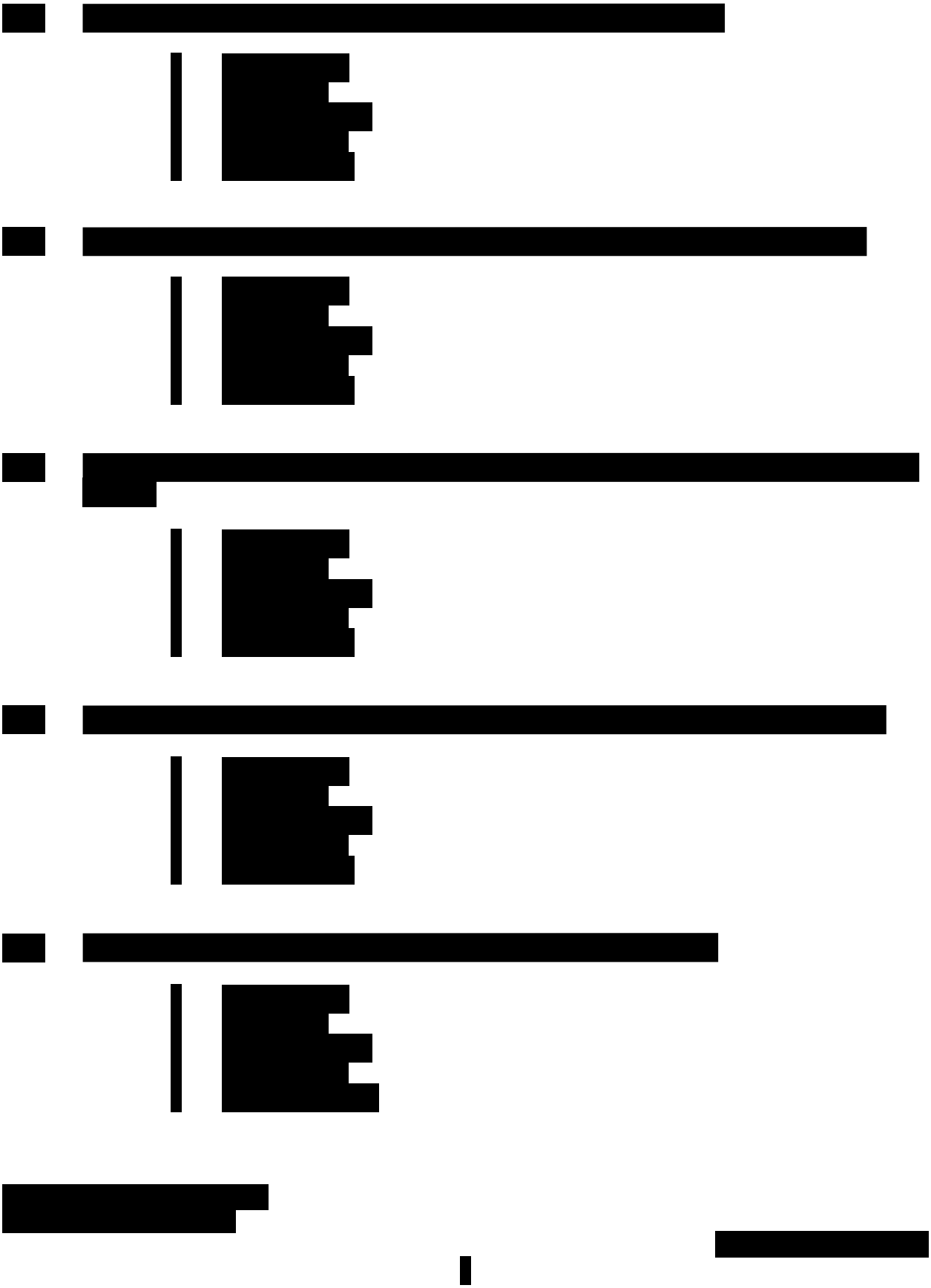


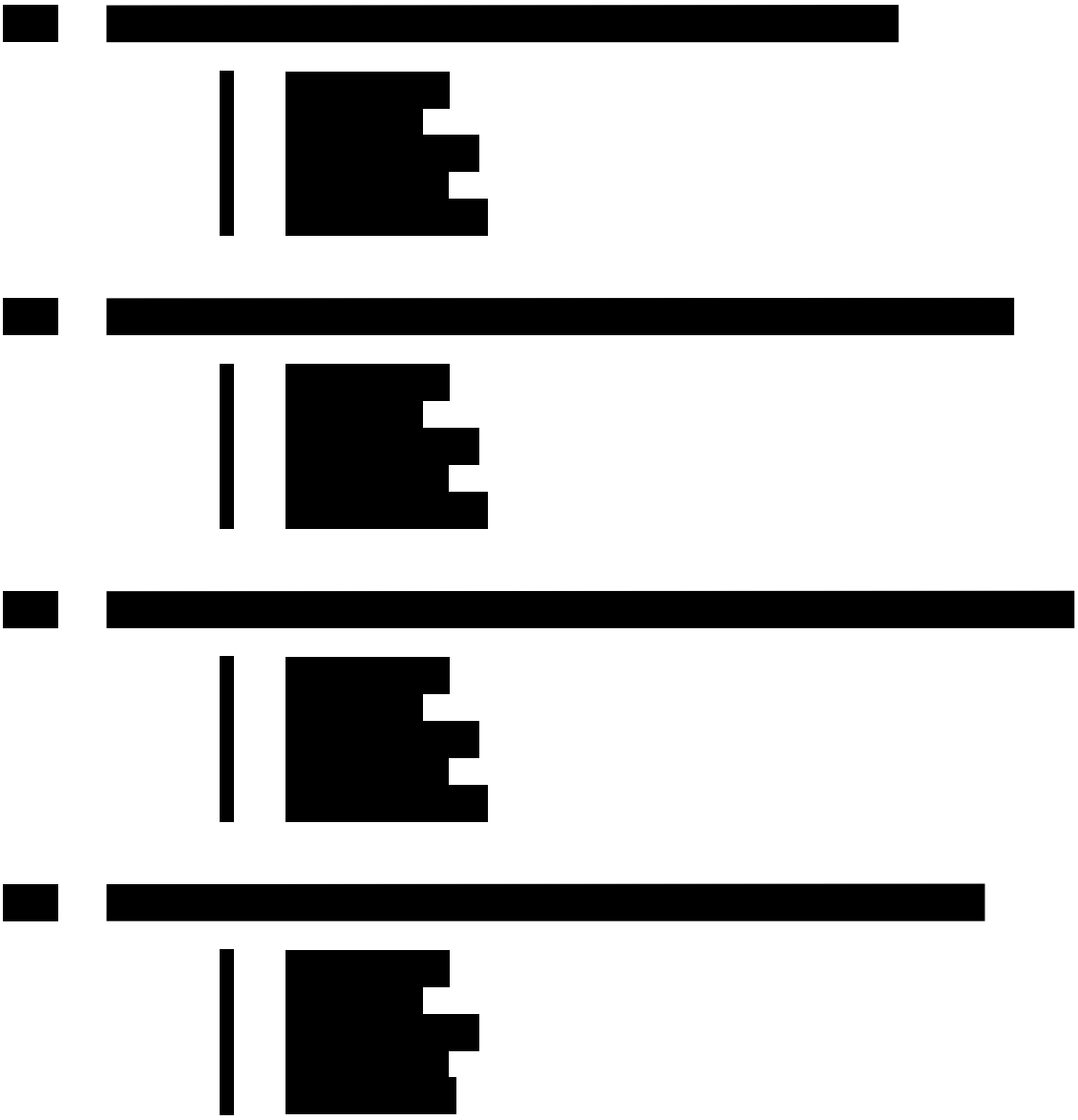
Source: Lewis SJ, Heaton KW. Scand J Gastroenterol (1997)

APPENDIX 5 IBS-QOL









APPENDIX 6 CDC HRQOL-4

Healthy Days Core Module (CDC HRQOL– 4)

1. Would you say that in general your health is

Please Read

a. Excellent 1
b. Very good 2
c. Good 3
d. Fair 4

or

e. Poor 5

Do not read these responses

Don't know/Not sure 7
Refused 9

2. Now thinking about your physical health, which includes physical illness and injury, for how many days during the past 30 days was your physical health not good?

a. Number of Days _ _
b. None 8 8

Don't know/Not sure 7 7
Refused 9 9

3. Now thinking about your mental health, which includes stress, depression, and problems with emotions, for how many days during the past 30 days was your mental health not good?

a. Number of Days _ _
b. None 8 8 If both Q2 AND Q3 = "None," skip next question

Don't know/Not sure 7 7
Refused 9 9

4. During the past 30 days, for about how many days did poor physical or mental health keep you from doing your usual activities, such as self-care, work, or recreation?

a. Number of Days _ _
b. None 8 8

Don't know/Not sure 7 7
Refused 9 9

APPENDIX 7 PRIOR IBS-C SYMPTOM MANAGEMENT ASSESSMENT

The Prior IBS-C Symptom Management Assessment will be incorporated within the patient's source documentation.

Prior IBS-C Symptom Management Assessment

This questionnaire is to be administered by the Coordinator

Treatment or Modification	Has the patient ever used this treatment for IBS-C? 1. Yes 2. No	If yes, when was the last time it was taken/used? 1. Currently using (record con meds in eCRF) 2. Less than 6 months ago 3. More than 6 months but less than 1 year ago 4. Over one year ago	If used, how satisfied was the patient with the treatment or modification's ability to relieve: Bowel symptoms (eg, constipation)? 1. Not at all satisfied 2. A little satisfied 3. Moderately satisfied 4. Quite satisfied 5. Very satisfied	If used, how satisfied was the patient with the treatment or modification's ability to relieve: Abdominal symptoms (eg, bloating, discomfort, pain)? 1. Not at all satisfied 2. A little satisfied 3. Moderately satisfied 4. Quite satisfied 5. Very satisfied	If the patient stopped taking this medication, what was the primary reason? 1. Not applicable, currently taking 2. Did not improve abdominal symptoms 3. Did not improve bowel symptoms 4. Experienced side effects 5. Costs too much 6. Other
Diet modification					
Lifestyle modification (eg, exercise, psychological therapy, meditation)					
PEG Laxatives (OTC)					
PEG Laxatives (prescription)					
Bulk Laxatives					
Stimulant Laxatives					
Stool Softeners*					
Linaclotide (GC-C agonist)					
Plecanatide (GC-C agonist)					
Lubiprostone (Chloride channel activator)					
Antispasmodics					
Tegaserod (5-HT4 agonist)					
Opioid Pain Medication					

Other (eg, probiotics, peppermint oil, antidepressants, other non-opioid pain medications, prucalopride [5-HT4 agonist], tenapanor [NHE3 inhibitor]) Write in: _____ _____ _____					

* Note: Combination products, including docusate with stimulant laxatives (either bisacodyl or Senna) should be recorded according to the stimulant laxative included in the product. If this is a docusate plus bisacodyl combination product, this should be recorded as a bisacodyl product. If this is a docusate plus Senna combination product, this should be recorded as a Senna product.
Examples of combination stool softener and laxative products include:
Senekot-S, Senna Plus, Senna S, Senosol-SS, Peri-Colace (all of these contain Senna)
Generic products would say Docusate plus Senna or bisacodyl

Treatment Examples:

PEG Laxatives
Some examples are:
MiraLax
Dulcolax Balance
Polyethylene Glycol (PEG)

Bulk Laxatives
Psyllium products
Some examples are:
Metamucil
Metamucil Sugar Free
Psyllium Husk
Wheat Dextrin products
Some examples are:
Benefiber
Wheat Dextrin

Stimulant Laxatives
Bisacodyl products
Some examples are:
Dulcolax
Bisacodyl
Ex-Lax Ultra Strength
Senna products
Some examples are:
Senna
Senakot
Ex-Lax Regular strength
Ex-Lax Maximum strength

Stool Softeners*
Docusate products
Some examples are:
Docusate sodium
Colace
Dulcolax Stool Softener

APPENDIX 8 INVESTIGATOR'S SIGNATURE

Study Title:	A Phase 3b, Randomized, Double-blind, Placebo-controlled, Parallel-group Trial of Linaclotide 290 µg Administered Orally for 12 Weeks Followed by a 4-week Randomized Withdrawal Period in Patients with Irritable Bowel Syndrome with Constipation
Study Number:	MCP-103-312
Final Date:	26 March 2018

I have read the protocol described above. I agree to comply with all applicable regulations and to conduct the trial as described in the protocol.

Signed:_____ Date:_____

Print Name:_____

15. REFERENCE LIST

1. Drossman DA, Corazziari E, Delvaux N, Spiller R, Talley NJ, Thompson CA, et al. Rome III: The functional gastrointestinal disorders. 3 ed. McLean, VA: Degnon Associates; 2006.

2. Sandler RS, Jordan MC, Shelton BJ. Demographic and dietary determinants of constipation in the US population. Am J Public Health. 1990;80(2):185-9.

3. Camilleri M, Prather CM. The irritable bowel syndrome: mechanisms and a practical approach to management. Ann Intern Med. 1992;116(12 Pt 1):1001-8.

4. Thompson WG. Irritable bowel syndrome: pathogenesis and management. Lancet. 1993;341(8860):1569-72.

5. Lynn RB, Friedman LS. Irritable bowel syndrome. N Engl J Med. 1993;329(26):1940-5.

6. Zighelboim J, Talley NJ. What are functional bowel disorders? Gastroenterology. 1993;104(4):1196-201.

7. Drossman DA, Camilleri M, Mayer EA, Whitehead WE. AGA technical review on irritable bowel syndrome. Gastroenterology. 2002;123(6):2108-31.

8. Longstreth GF, Thompson WG, Chey WD, Houghton LA, Mearin F, Spiller RC. Functional bowel disorders. Gastroenterology. 2006;130:1480-91.

9. Forte LR. Guanylin regulatory peptides: structures, biological activities mediated by cyclic GMP and pathobiology. Regul Pept. 1999;81(1-3):25-39.

10. Sindic A, Schlatter E. Cellular Effects of Guanylin and Uroguanylin. J Am Soc Nephrol. 2005;17(3):607-16.

11. Castro J, Harrington AM, Hughes PA, Martin CM, Ge P, Shea CM, et al. Linaclotide Inhibits Colonic Nociceptors and Relieves Abdominal Pain via Guanylate Cyclase-C and Extracellular Cyclic GMP. Gastroenterology. 2013;145(6):1334-46.

12. Chey WD, Lembo AJ, Lavins BJ, Shiff SJ, Kurtz CB, Currie MG, et al. Linaclotide for Irritable Bowel Syndrome With Constipation: A 26-Week, Randomized, Double-blind, Placebo-Controlled Trial to Evaluate Efficacy and Safety. The American Journal of Gastroenterology. 2012;107(11):1702-12.

13. Rao S, Lembo AJ, Shiff SJ, Lavins BJ, Currie MG, Jia XD, et al. A 12-week, randomized, controlled trial with a 4-week randomized withdrawal period to evaluate the efficacy and safety of linaclotide in irritable bowel syndrome with constipation. Am J Gastroenterol. 2012;107(11):1714-24; quiz p.25.

14. Lewis SJ, Heaton KW. Stool form scale as a useful guide to intestinal transit time. Scand J Gastroenterol. 1997;32(9):920-4.

15. Patrick DL, Drossman DA, Frederick IO, DiCesare J, Puder KL. Quality of life in persons with irritable bowel syndrome: development and validation of a new measure. Dig Dis Sci. 1998;43(2):400-11.

16. Prevention CfDCa. Measuring Healthy Days: Population Assessment of Health-Related Quality of Life. Centers for Disease Control and Prevention [Internet]. 2000. Available from: https://www.cdc.gov/hrqol/pdfs/mhd.pdf.

The data and information related to my line function, which has been included with this file, are truthful and accurate.

Approval	[redacted]
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