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(54) Titre : PROCEDES DE TRAITEMENT DU SYNDROME DU COLON IRRITABLE AVEC DES FORMULATIONS A
LIBERATION MODIFIEE OU RETARDEE DE LINACLOTIDE
(54) Title: TREATMENT METHODS AND DELAYED RELEASE COMPOSITION OF LINACLOTIDE

(57) **Abrégé/Abstract:**

The invention provides methods for treating a patient with a disorder, such as a GI disorder or symptoms associated with a GI or non-GI disorder, by administering a therapeutically effective amount of a delayed release pharmaceutical composition comprising linacotide.

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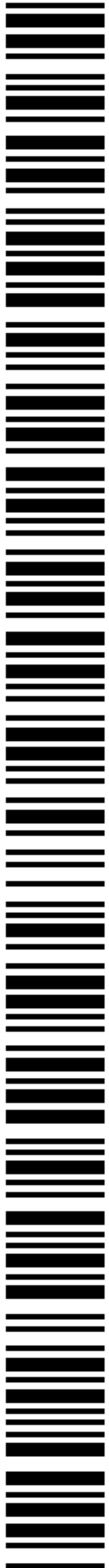
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(54) Title: METHODS OF TREATING IRRITABLE BOWEL SYNDROME WITH MODIFIED OR DELAYED RELEASE FOR-
MULATIONS OF LINACLOTIDE

(57) Abstract: The invention provides methods for treating a patient with a disorder, such as a GI disorder or symptoms associated
with a GI or non-GI disorder, by administering a therapeutically effective amount of a delayed release pharmaceutical composition
comprising linaclotide.

TREATMENT METHODS WITH DELAYED RELEASE COMPOSITION OF LINACLOTIDE

FIELD OF THE INVENTION

[0001] The invention relates to the use of delayed release pharmaceutical compositions comprising linaclotide to treat a variety of indications, including gastrointestinal (GI) disorders, such as irritable bowel syndrome with constipation (IBS-c) and symptoms associated with GI or non-GI disorders, such as abdominal pain.

PRIORITY CLAIM

[0002] This application claims priority under 35 U.S.C. § 119(e) to U.S. Provisional Patent Application Number 62/437,566 filed on December 21, 2016, the entire contents of which are hereby incorporated by reference.

SEQUENCE LISTING

[0003] This application incorporates by reference in its entirety the Sequence Listing entitled "IW196PCT1_ST25.txt" (4,514 bytes) which was created on December 20, 2017 and filed electronically herewith.

BACKGROUND OF THE INVENTION

[0004] Tens of millions of adults in the U.S. suffer from gastrointestinal (GI) disorders, such as irritable bowel syndrome (IBS), chronic idiopathic constipation (CIC), diverticulitis, and ulcerative colitis. Many others suffer from symptoms such as abdominal pain, discomfort and bloating associated with GI or non-GI disorders. An estimated 13 million adults in the U.S. suffer from irritable bowel syndrome with constipation (IBS-c). IBS-c is a chronic functional gastrointestinal disorder characterized by abdominal pain, discomfort, and bloating associated with altered bowel habits. There are currently few available therapies to treat these disorders and there is a high rate of dissatisfaction with them. Patients suffering from GI disorders, such as IBS-c can be affected physically, psychologically, socially, and economically.

[0005] Rome III Diagnostic Criteria for irritable bowel syndrome (IBS) includes recurrent abdominal pain or discomfort more than 3 days per month over the previous 3 months before the diagnosis, with the onset at least 6 months before the diagnosis, and associated with two or more of the following:

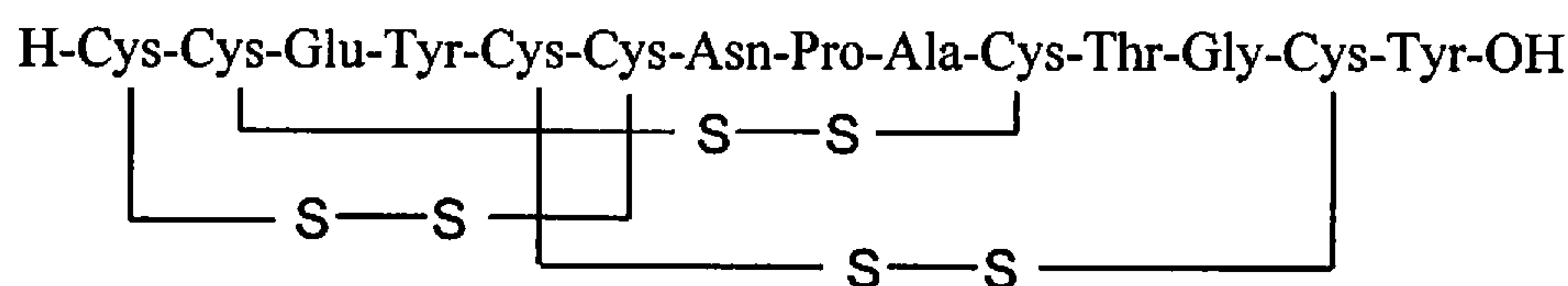
- a.* The abdominal discomfort or pain is relieved with defecation;

- b.* Onset of symptoms is associated with a change in frequency of stools;
- c.* Onset of symptoms is associated with a change in the form or appearance of stool.

[0006] Rome III Diagnostic Criteria further defines irritable bowel syndrome with constipation (IBS-c) in a patient as having hard or lumpy stool (Bristol Stool Form Scale 1-2) with at least 25% of BMs and loose or watery stool (Bristol Stool Form Scale 6-7) with less than 25% of BMs.

[0007] Patients with IBS-c may also report symptoms that include (i) alternation between constipation and normal stools, and (ii) lower abdominal cramping, aching or discomfort that is commonly triggered by eating.

[0008] U.S. Patents 7,304,036 and 7,371,727, herein incorporated by reference, disclose peptides that act as agonists of the guanylate cyclase C (GC-C) receptor for the treatment of gastrointestinal (GI) disorders. One particular peptide disclosed is linaclotide, which consists of the following amino acid sequence: Cys Cys Glu Tyr Cys Cys Asn Pro Ala Cys Thr Gly Cys Tyr. Linaclotide has the chemical structure of:



[0009] Linaclotide is orally administered and has been approved in the U.S. by the FDA for the treatment of irritable bowel syndrome with constipation (IBS-c) and chronic idiopathic constipation (CIC). In humans, linaclotide has been shown to affect GI physiology including reducing visceral pain, reducing bloating and accelerating GI transit which can lead to increased stool frequency and improved stool consistency. Orally administered linaclotide acts locally by binding to and activating GC-C receptors at the luminal surface of the intestine. The GC-C receptor is a key regulator in mammals of intestinal function and is found throughout the luminal surface of the GI tract. The GC-C receptor responds to the endogenous hormones, guanylin and uroguanylin, and to enteric bacterial peptides from the heat stable enterotoxin family (ST peptide). When linaclotide binds to the GC-C receptor, there is an intracellular elevation of the second messenger, cyclic GMP (c-GMP), and an increase in chloride and bicarbonate secretion, resulting in an increase in intestinal fluid secretion and reducing abdominal pain.

[00010] As approved by the FDA, linaclotide is administered in an oral, solid, immediate-release capsule formulation manufactured by filling drug-layered beads into gelatin capsules. Due to the high expression of GC-C receptors throughout GI tract, linaclotide from

an immediate release formulation activates the GC-C receptor starting from the upper GI tract, resulting in significant amount of intestinal fluid being brought to the lower GI tract. To reduce or mitigate this effect, delayed release compositions are needed which have targeted release of linaclotide in the distal or lower segment of the gastrointestinal tract. Targeting the lower GI for linaclotide release may help avoid excess fluid secretion but at the same time maintain or improve linaclotide efficacy for treating abdominal and bowel symptoms of GI disorders.

[00011] Various formulation techniques have been used to develop delayed release compositions for pharmaceutically active agents including the use of enteric coatings or pH responsive polymers. However, the specific components of these compositions vary greatly and depend significantly on the particular pharmaceutically active agent and the desired properties. For example, the formulation must be compatible with the pharmaceutically active agent and also provide the necessary dissolution performance and stability properties.

[00012] Linaclotide has been previously demonstrated to reduce visceral pain in the GI tract which is thought to be mediated by increasing cGMP. Animal studies have shown that orally administered linaclotide can treat colonic hypersensitivity and hyperalgesia. However, due to the reducing environment of the intestinal tract, it is known that much of the oral linaclotide dose is degraded prior to reaching the distal colon. In human volunteers treated with linaclotide, only about 5% of the oral dose is found in feces. Delayed release (“DR”) compositions of linaclotide that target the lower GI may improve linaclotide’s efficacy towards relieving pain associated with various GI disorders by allowing for delivery of a higher dose of linaclotide to the colon. Such DR compositions of linaclotide would have the potential to release linaclotide predominantly (or fully) in the lower GI. As a result, for example, the DR formulation or composition may have an increased capacity to treat lower GI associated disorders. Surprisingly, orally administered linaclotide has also been demonstrated to reduce visceral pain in non-GI tissues, providing further evidence that the mechanism of visceral pain relief via linaclotide is not mediated solely by promoting secretion. This result suggests that a cGMP modulator whose distribution is limited to the GI can relieve pain can be used as a therapy to relieve pain in other parts of the body. However, in order for linaclotide to be a useful therapy for the treatment of visceral pain in non-GI tissues (e.g., ulcerative colitis, diverticulitis, IBS, overactive bladder syndrome, bladder hypersensitivity or colitis induced bladder afferent hyperactivity, etc.) for non-constipated patients, it would be necessary to reduce or eliminate the secretion-promoting effects of linaclotide. As such, in one aspect, there is a need to develop a means of at least partially, or completely separating the effect of linaclotide in promoting secretion from that of relieving visceral pain.

[00013] Moreover, it may have a capacity to cause lower incidences of adverse events (such as diarrhea) than the immediate-release dosage form, e.g., because it would cause lower overall intestinal fluid secretion by not activating GC-C receptors in the upper GI. This would occur while maintaining or even improving linaclotide efficacy for treating symptoms of GI disorders, such as pain.

[00014] Despite the need for delayed release compositions of linaclotide, difficulties exist in preparing such formulations due to the intrinsic chemical instability of linaclotide, for example, by moisture induced degradation reactions such as hydrolysis, deamidation, isomerization, and multimerization. These difficulties may be exacerbated when producing formulations having lower dosages of linaclotide.

[00015] Accordingly, there is a need for delayed release compositions that provide stable and reliable delivery of linaclotide to targeted areas of the gastrointestinal tract. There is also a need for methods of treating IBS-c by administering delayed release compositions of linaclotide.

SUMMARY OF THE INVENTION

[00016] In general, the invention relates to a method of treating disorders, such as gastrointestinal (GI) disorders (e.g., IBS-c) or symptoms associated with GI or non-GI disorders (e.g., abdominal pain).

[00017] One aspect of the invention is a method of reducing the incidence or severity of an adverse event associated with administration of linaclotide, comprising orally administering to a subject a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet comprises an enteric coating comprising a pH-sensitive polymer that releases linaclotide in a lower GI of the subject when compared to administration of an equal dose of an immediate release dosage form of linaclotide, and the delayed-release pharmaceutical tablet composition further comprises a therapeutically effective amount of linaclotide to treat constipation and/or pain in the subject.

[00018] Another aspect of the invention is a method of reducing the incidence or severity of an adverse event associated with administration of linaclotide in a subject, comprising orally administering a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet comprises an enteric coating comprising a pH-sensitive polymer that releases a therapeutically effective amount of linaclotide to treat constipation and/or pain in the subject.

[00019] Another aspect of the invention is a method of reducing intestinal fluid secretion-promoting effects of linaclotide, comprising orally administering to a subject a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet

further comprises an enteric coating comprising a pH-sensitive polymer that releases linaclotide in a lower GI of the subject.

[00020] Yet another aspect of the invention is a method of treating visceral or abdominal pain in a non-constipated subject, comprising orally administering a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet further comprises an enteric coating and a pH sensitive polymer that releases an amount of linaclotide in a stomach of the subject that is less than an amount effective to induce secretion in the subject.

[00021] Another aspect of the invention is a method of treating irritable bowel syndrome with constipation (IBS-c) comprising orally administering to a patient in need thereof, a delayed release pharmaceutical tablet composition comprising a therapeutically effective amount of linaclotide.

[00022] Still another aspect of the invention is a method of treating abdominal pain comprising orally administering to a patient in need thereof, a delayed release pharmaceutical tablet composition comprising a therapeutically effective amount of linaclotide.

BRIEF DESCRIPTION OF THE FIGURES

[00023] Figure 1 is an illustration of three distinct periods of the trial described in Example 18.

[00024] Figure 2 is a plot of the primary efficacy parameter, 6/12 week APC +1 responder, for the eight treatment groups as described in Example 20.

[00025] Figures 3A and 3B are a plot of the secondary efficacy parameter, weekly SBM frequency over baseline, for the eight treatment groups as described in Example 20.

[00026] Figure 4 is a plot of the primary efficacy parameter, weekly abdominal pain change from baseline, for the eight treatment groups as described in Example 20.

[00027] Figure 5 is a plot of the secondary efficacy parameter, 12-week CSBM frequency over baseline, for the eight treatment groups as described in Example 20.

[00028] Figures 6A and 6B are a plot of the secondary efficacy parameters, 6/12 and 9/12 CSBM responder, for the eight treatment groups as described in Example 20.

[00029] Figures 7A and 7B are a plot of the secondary efficacy parameters, 6/12 and 9/12 abdominal pain responder, for the eight treatment groups as described in Example 20.

[00030] Figure 8 is a plot of the secondary efficacy parameter, 12-week abdominal pain change from baseline, for the eight treatment groups as described in Example 20.

[00031] Figure 9 is a plot of the secondary efficacy parameter, 12-week SBM frequency over baseline, for the eight treatment groups as described in Example 20.

[00032] Figure 10 is a plot of the secondary efficacy parameter, weekly SBM frequency over baseline, for the eight treatment groups as described in Example 20.

[00033] Figure 11 is a plot of the secondary efficacy parameter, 12-week stool consistency (BSFS) change from baseline, for the eight treatment groups as described in Example 20.

[00034] Figure 12 is a plot of the secondary efficacy parameters, 12-week abdominal symptom score change over baseline, 12-week abdominal pain change over baseline, 12 week abdominal discomfort change over baseline, and 12-week abdominal bloating change over baseline, for the eight treatment groups as described in Example 20.

[00035] Figure 13 is a plot of the secondary efficacy parameter, treatment satisfaction, for the eight treatment groups as described in Example 20.

[00036] Figure 14 is a plot of overall diarrhea TEAE rates for the eight treatment groups as described in Example 20.

[00037] Figure 15 is a plot of the overall diarrhea and diarrhea leading to drop out (ADO) rates for the eight treatment groups as described in Example 20.

[00038] Figure 16 is a plot of diarrhea TEAEs by severity for the eight treatment groups as described in Example 20.

[00039] Figure 17A, Figure 17B and Figure 17C is a plot of certain efficacy parameters compared with data from the Phase III clinical trial for Linzess®.

[00040] Figure 18 is a plot of the diarrhea TEAE rates compared with data from the Phase III clinical trial for Linzess®.

[00041] Figure 19 is a plot of the diarrhea TEAEs by severity compared with data from the Phase III clinical trial for Linzess®.

[00042] Figure 20A shows the release profile at various pHs for delayed release compositions comprising 100µg linaclotide. Figure 20 B shows the effect of altering the ratios of pH-sensitive polymers on release profiles.

[00043] Figure 21 is a plot of the percent change from baseline in weekly abdominal pain at week 12 for the DR1 composition, the IR composition and placebo.

DETAILED DESCRIPTION OF THE INVENTION

[00044] *A. Methods of Treatment*

[00045] In one aspect, described generally herein are methods of treatment comprising orally administering a delayed release composition comprising linaclotide, which are used to treat any number of diseases, disorders or symptoms involving constipation and/or pain (e.g., visceral pain, abdominal pain). For example, the methods of treatment comprising orally

administering a delayed release pharmaceutical tablet composition comprising linaclotide, as described herein, are used to treat irritable bowel syndrome with constipation (IBS-c) in a patient in need thereof. The patient may be diagnosed with IBS-c according to the Rome Criteria (e.g. Rome III). In another embodiment, the methods of treatment comprising orally administering a delayed release pharmaceutical tablet composition comprising linaclotide, as described herein, are used to treat abdominal pain in a patient in need thereof.

[00046] One aspect of the invention is a method of reducing the incidence or severity of an adverse event associated with administration of linaclotide, comprising orally administering to a subject a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet comprises an enteric coating comprising a pH-sensitive polymer that releases linaclotide in a lower GI of the subject when compared to administration of an equal dose of an immediate release dosage form of linaclotide, and the delayed-release pharmaceutical tablet composition further comprises a therapeutically effective amount of linaclotide to treat constipation and/or pain in the subject.

[00047] Another aspect of the invention is a method of reducing the incidence or severity of an adverse event associated with administration of linaclotide in a subject, comprising orally administering a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet comprises an enteric coating comprising a pH-sensitive polymer that releases a therapeutically effective amount of linaclotide to treat constipation and/or pain in the subject.

[00048] In some embodiments, the reduction in the incidence or severity of the adverse event is a reduction in the incidence or severity of the adverse event compared to administration of an equal dose of an immediate release dosage form of linaclotide. In some embodiments, the adverse event is diarrhea. In some embodiments, the incidence of the adverse event is reduced. In some embodiments, the severity of the adverse event is reduced.

[00049] Another aspect of the invention is a method of reducing intestinal fluid secretion-promoting effects of linaclotide, comprising orally administering to a subject a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet further comprises an enteric coating comprising a pH-sensitive polymer that releases linaclotide in a lower GI of the subject.

[00050] Yet another aspect of the invention is a method of treating visceral or abdominal pain in a non-constipated subject, comprising orally administering a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet further comprises an enteric coating and a pH sensitive polymer that releases an amount of linaclotide in a stomach of the subject that is less than an amount effective to induce secretion in the subject.

[00051] Another aspect of the invention is a method of treating irritable bowel syndrome with constipation (IBS-c) comprising orally administering to a patient in need thereof, a delayed release pharmaceutical tablet composition comprising a therapeutically effective amount of linaclotide.

[00052] Still another aspect of the invention is a method of treating abdominal pain comprising orally administering to a patient in need thereof, a delayed release pharmaceutical tablet composition comprising a therapeutically effective amount of linaclotide.

[00053] In some embodiments, the delayed-release pharmaceutical tablet composition comprises a therapeutically effective amount of linaclotide to reduce, prevent or relieve pain or constipation in the subject. In some embodiments, the delayed-release pharmaceutical composition comprises a therapeutically effective amount of linaclotide to reduce, prevent or relieve pain in the subject, but does not affect bowel habit. In some embodiments, the delayed-release pharmaceutical composition provides less than an amount of linaclotide effective to substantially affect bowel habit. In some embodiments, the bowel habit is selected from CSBM rate, SBM rate, or stool consistency.

[00054] In some embodiments, the subject is diagnosed with irritable bowel syndrome with constipation (IBS-c).

[00055] In some embodiments, the delayed-release pharmaceutical tablet composition comprising linaclotide releases less than 50%, less than 40%, less than 25% or less than 10% of the linaclotide in the stomach.

[00056] In some embodiments, the delayed release pharmaceutical tablet composition is administered once daily. In some embodiments, the delayed release pharmaceutical tablet composition is administered once daily in the morning. In some embodiments, the delayed release pharmaceutical tablet composition is administered once daily in the morning at least 30 minutes before breakfast. In some embodiments, the delayed release pharmaceutical tablet composition is administered after the patient has fasted for at least 2 hours. In some embodiments, the delayed release pharmaceutical tablet composition is administered for at least 12 weeks.

[00057] In some embodiments, the administering increases the number of complete spontaneous bowel movements (CSBMs) by the patient. In some embodiments, the administering decreases abdominal pain in the patient. In some embodiments, the administering increases the number of complete spontaneous bowel movements (CSBMs) by the patient from baseline by at least one per week during at least 6 out of 12 weeks. In some embodiments, the administering decreases the abdominal pain in the patient from baseline by at least 30% during at least 6 out of 12 weeks. In some embodiments, The method of any of

the preceding claims, wherein the administering increases the number of complete spontaneous bowel movements (CSBMs) by the patient from baseline by at least one per week during at least 6 out of 12 weeks; and wherein the administering decreases the abdominal pain in the patient from baseline by at least 30% during at least 6 out of 12 weeks.

[00058] In some embodiments, the administering improves one or more (e.g., two or more) of the following: abdominal pain, CSBM frequency rate, SBM frequency rate, stool consistency, straining, abdominal discomfort, abdominal bloating, abdominal symptom score, constipation severity, IBS symptom severity, days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

[00059] In some embodiments, the administering results in improvement in one or more (e.g., two or more) of the following efficacy parameters: weekly change from baseline in abdominal pain, weekly change from baseline in CSBM frequency, 6/12 week APC+1 responder, 6/12 week CSBM +1 responder, 9/12 week CSBM +1 responder, 6/12 week abdominal pain responder, 9/12 week abdominal pain responder, weekly CSBM +1 responder, weekly abdominal pain responder, weekly APC +1 responder, change from baseline in 12-week abdominal pain, change from baseline in 12-week CSBM frequency rate, change from baseline in 12-week SBM frequency rate, change from baseline in 12-week stool consistency, change from baseline in 12-week straining, change from baseline in 12-week abdominal discomfort, change from baseline in 12-week abdominal bloating, change from baseline in 12-week abdominal symptom score, change from baseline in 12-week constipation severity, change from baseline in 12-week IBS symptom severity, change from baseline in 12-week percent of days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

[00060] In some embodiments, the administering improves, as compared to treatment with an immediate release composition of linaclotide, one or more of the following: abdominal pain, CSBM frequency rate, SBM frequency rate, stool consistency, straining, abdominal discomfort, abdominal bloating, abdominal symptom score, constipation severity, IBS symptom severity, days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

[00061] In some embodiments, the administering results in improvement, as compared to treatment with an immediate release composition of linaclotide, in one or more of the following efficacy parameters: weekly change from baseline in abdominal pain, weekly change from baseline in CSBM frequency, 6/12 week AP+1 responder, 6/12 week CSBM +1 responder, 9/12 week CSBM +1 responder, 6/12 week abdominal pain responder, 9/12 week abdominal pain responder, weekly CSBM +1 responder, weekly abdominal pain responder,

weekly APC +1 responder, change from baseline in 12-week abdominal pain, change from baseline in 12-week CSBM frequency rate, change from baseline in 12-week SBM frequency rate, change from baseline in 12-week stool consistency, change from baseline in 12-week straining, change from baseline in 12-week abdominal discomfort, change from baseline in 12-week abdominal bloating, change from baseline in 12-week abdominal symptom score, change from baseline in 12-week constipation severity, change from baseline in 12-week IBS symptom severity, change from baseline in 12-week percent of days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

[00062] In some embodiments, the administering results in a decrease in the risk of adverse events in the patient compared to treatment with an immediate release composition of linaclotide. In some embodiments, the administering results in a decrease in the risk of diarrhea in the patient compared to treatment with an immediate release composition of linaclotide. In some embodiments, the administering results in a decrease of at least 20% in the risk of diarrhea in the patient compared to treatment with an immediate release composition of linaclotide. In some embodiments, the administering results in a decrease in the risk of severe diarrhea in the patient compared to treatment with an immediate release composition of linaclotide.

[00063] Another aspect of the invention is a method of treating or relieving pain comprising administering to a patient in need thereof, a therapeutically effective amount of a delayed-release pharmaceutical tablet composition as described herein.

[00064] In some embodiments, the pain is selected from visceral pain; diverticulitis pain; pelvic pain; abdominal pain; or pain associated with gastrointestinal disorders, venereal diseases, bladder pain syndrome, or interstitial cystitis. In some embodiments, the pain is selected from general abdominal pain, diverticular disease, pain associated with irritable bowel syndrome (IBS), chronic or acute radiation proctopathy (also referred to as radiation proctitis), rectal pain, chronic proctalgia, proctalgia fugax, anal pain, chronic anal fissure, post-operative anal pain, overactive bladder syndrome, stress incontinence, interstitial cystitis, bladder pain syndrome, pain associated with cancer, pain associated with gastrointestinal tract neoplasms, general pelvic pain, endometriosis, orchialgia, chronic prostatitis, prostatodynia, vulvodynia, urethral syndrome, penile pain, perianal pain, and pain associated with ulcerative colitis, ulcerative proctitis, or Crohn's disease.

[00065] In some embodiments, the method of treating a patient includes administering a delayed release composition comprising a therapeutically effective amount of linaclotide once a day. In some embodiments, the composition is administered once a day in the morning. In some embodiments, the composition is administered once a day at least 30 minutes before ingestion of food. For example, once a day in the morning at least 30 minutes before breakfast.

In some embodiments, the composition is administered after the patient has fasted, e.g., after the patient has fasted for at least 2 hours, for at least 4 hours, for at least 8 hours, or for at least 10 hours.

[00066] In certain aspects of the present methods, the composition is administered for a period of greater than four weeks, (e.g., at least 8 weeks, at least 12 weeks, or at least 26 weeks). In some aspects of the present method, the linaclotide is administered each day of the week, at least once a week, at least twice a week, at least three times a week, at least four times a week, at least five times a week or at least six times a week.

[00067] In one aspect, the method of treating a patient includes orally administering a delayed release composition comprising a therapeutically effective amount of linaclotide, wherein the administering increases the frequency of complete spontaneous bowel movements (CSBMs) by the patient. In some embodiments, the CSBM frequency rate is increased to three or more CSBMs per week. In some embodiments, the CSBM frequency rate is increased by one or more CSBMs per week compared to a baseline level of CSBMs prior to treatment with delayed release compositions of linaclotide. In some embodiments, the frequency of CSBMs by the patient is increased compared to treatment with immediate release compositions of linaclotide. In some aspects, the administering increases the spontaneous bowel movement (SBM) frequency rate for the patient. In some embodiments, the frequency of SBMs is increased compared to treatment with immediate release compositions of linaclotide.

[00068] In another aspect, the method of treating a patient includes administering a delayed release composition comprising a therapeutically effective amount of linaclotide, wherein the administering decreases abdominal pain in the patient. In some embodiments, the abdominal pain is decreased by at least 30% (e.g., at least 40%, at least 50%) compared to a baseline level of abdominal pain prior to treatment with delayed release compositions of linaclotide. In some embodiments, abdominal pain in the patient is decreased compared to treatment with immediate release compositions of linaclotide. In some embodiments, the abdominal pain is decreased by at least at least 30% (e.g., at least 40%, at least 50%) at week 12, after 12 weeks of administration.

[00069] In another aspect, the method of treating a patient includes administering a delayed release composition comprising a therapeutically effective amount of linaclotide, wherein the administering increases the frequency of complete spontaneous bowel movements (CSBMs) by the patient and decreases abdominal pain in the patient.

[00070] In another aspect, the method of treating a patient includes administering a delayed release composition comprising a therapeutically effective amount of linaclotide, wherein the administering improves abdominal symptoms (e.g., pain, discomfort, bloating)

and/or bowel symptoms (e.g., CSBMs/per week, SBMs/per week, stool consistency, straining) in a patient with greater than moderate abdominal pain, in a patient with moderate to severe abdominal pain, or in a patient with severe to very severe abdominal pain.

[00071] Importantly, administration of delayed release compositions comprising linaclotide as described herein may provide one or more of the following advantages: an increase in the number of CSBMs, a decrease in abdominal pain or discomfort, a decrease in bloating, a decrease in constipation severity, an improvement in stool consistency, an increase in SBM frequency, an increase in BM frequency, a decrease in straining during defecation, an improvement in abdominal symptom score, an improvement in IBS symptom severity, a decrease in use of per-protocol rescue medicine, and an improvement in patient assessment of quality of life, treatment satisfaction or adequate relief. The improvement may be an improvement as compared to treatment with immediate release composition of linaclotide.

[00072] In some embodiments, the method of treating a patient includes treating a disorder selected from irritable bowel syndrome (IBS), constipation, irritable bowel syndrome with constipation (IBS-c), irritable bowel syndrome with diarrhea (IBS-d), mixed IBS (IBS-m), un-subtyped IBS (IBS-u), chronic idiopathic constipation (CIC), colon cancer, diverticulitis, ulcerative colitis, a functional gastrointestinal disorder, gastroesophageal reflux disease, functional heartburn, dyspepsia, visceral pain, abdominal pain, gastroparesis, chronic intestinal pseudo-obstruction, colonic pseudo-obstruction, Crohn's disease, inflammatory bowel disease, overactive bladder syndrome, bladder hypersensitivity or colitis induced bladder afferent hyperactivity in a patient in need thereof. In some embodiments, the method of treating a patient includes treating a symptom associated with a disorder, such as a GI disorder, or alternatively a non-GI disorder in a patient in need thereof. For example, the treatment may be for abdominal pain, discomfort or bloating, or visceral pain associated with a disorder (GI or non-GI). For example, the patient may be a non-constipated patient.

[00073] In further embodiments, the disorder or symptom being treated is selected from visceral pain; diverticulitis pain; pelvic pain; abdominal pain; or pain associated with gastrointestinal disorders, venereal diseases, bladder pain syndrome, or interstitial cystitis. In further embodiments, the disorder or symptom being treated is pain selected from general abdominal pain, diverticular disease, pain associated with irritable bowel syndrome (IBS), chronic or acute radiation proctopathy (also referred to as radiation proctitis), rectal pain, chronic proctalgia, proctalgia fugax, anal pain, chronic anal fissure, post-operative anal pain, overactive bladder syndrome, stress incontinence, interstitial cystitis, bladder pain syndrome, pain associated with cancer, pain associated with gastrointestinal tract neoplasms, general pelvic pain, endometriosis, orchialgia, chronic prostatitis, prostatodynia, vulvodynia, urethral

syndrome, penile pain, perianal pain, and pain associated with ulcerative colitis, ulcerative proctitis, or Crohn's disease. In further embodiments, the disorder or symptom being treated is a disorder or symptom related to constipation, such as chronic constipation, opioid induced constipation, post-surgical constipation (post-operative ileus), and constipation associated with neuropathic disorders or a combination of symptoms thereof (such as a combination of irritable bowel syndrome and chronic constipation), constipation associated with neuropathic disorders (e.g., constipation associated with Parkinson's Disease), constipation associated with cystic fibrosis or thyroid disease.

[00074] In some embodiments, the disorder or symptom being treated is a disorder or symptom associated with the lower GI (e.g., a lower GI disorder).

[00075] In some embodiments, the methods of treatment described herein are useful for the treatment of diseases or symptoms associated with visceral pain selected from the group consisting of general abdominal pain, diverticular disease, pain associated with irritable bowel syndrome (IBS), chronic or acute radiation proctopathy (also referred to as radiation proctitis), rectal pain, chronic proctalgia, proctalgia fugax, anal pain, chronic anal fissure, post-operative anal pain, overactive bladder syndrome, stress incontinence, interstitial cystitis, bladder pain syndrome, pain associated with cancer, pain associated with gastrointestinal tract neoplasms, general pelvic pain, endometriosis, orchialgia, chronic prostatitis, prostatodynia, vulvodynia, urethral syndrome, penile pain, perianal pain, ulcerative colitis, ulcerative proctitis, and Crohn's disease. In one particular embodiment, the compositions described herein are useful for the treatment of bladder pain syndrome. In another particular embodiment, the compositions described herein are useful for the treatment of overactive bladder syndrome (including for example bladder hypersensitivity or colitis induced bladder afferent hyperactivity).

[00076] In still another embodiment, the methods of treatment described herein are useful for the treatment of interstitial cystitis. In still another embodiment, the compositions described herein are useful for the treatment of endometriosis. In another embodiment, the compositions described herein are useful for the treatment of anal pain.

[00077] In some embodiments, a method of treating a disorder is provided comprising administering to a patient in need thereof, a therapeutically effective amount of the composition described herein. In some embodiments, the disorder is cancer selected from colorectal/local metastasized colorectal cancer, intestinal polyps, Barrett's esophagus, gastrointestinal tract cancer, lung cancer, cancer or pre-cancerous growths or metastatic growths of epithelial cells, polyps, breast, colorectal, lung, ovarian, pancreatic, prostatic, renal, stomach, bladder, liver, esophageal and testicular carcinoma.

[00078] In some embodiments, methods are provided for preventing a cancer or hyperplasia of the gastrointestinal tract or preventing reoccurrence of cancer or hyperplasia of the gastrointestinal tract in a patient in need thereof comprising administering an effective amount of the composition or the oral dosage form to the patient. In some embodiments, the cancer or hyperplasia is colorectal cancer, intestinal polyps or pre-cancerous growths or metastatic growths of gastrointestinal epithelial cells. In some embodiments, the composition or oral dosage form is administered simultaneously or sequentially with an effective amount of a COX-2 inhibitor. Examples of highly selective and selective COX-2 inhibitors include etoricoxib, rofecoxib, lumiracoxib, valdecoxib, celecoxib (Celebrex®), sulindac, diclofenac, meloxicam and etodolac. Non-selective NSAIDs that inhibit COX-2 include naproxen, ibuprofen, sodium salicylate and diflunisal. As used herein, the term “prevent” or “preventing” means to arrest, delay the onset (i.e., the period prior to clinical manifestation of a disease) or reoccurrence of cancer or hyperplasia, and/or reduce the risk of developing cancer or hyperplasia relative to a patient that has not been treated with a composition described herein.

[00079] In some embodiments, methods are provided for treating gastrointestinal disorders in pediatric patients with the compositions and oral dosage forms described herein. In some embodiments, methods are provided for treating gastrointestinal disorders in a pediatric patient diagnosed with one or more gastrointestinal disorders or conditions, wherein the method comprises administering an effective amount of the composition or the oral dosage form to the patient. In some embodiments, methods are provided to use the compositions and oral dosage forms for treating gastrointestinal disorders including, but not limited to, GI motility disorders, irritable bowel syndrome, constipation-predominant irritable bowel syndrome (IBS-c), mixed-type irritable bowel syndrome (IBS-m), chronic constipation, chronic idiopathic constipation, opioid induced constipation, post-surgical constipation (post-operative ileus), constipation associated with neuropathic disorders, constipation associated with cystic fibrosis or thyroid disease, dyspepsia (including functional dyspepsia or non-ulcer dyspepsia), gastroparesis, gastrointestinal motility disorders, functional gastrointestinal disorders, gastroesophageal reflux disease (GERD), inflammatory bowel disease, Crohn's disease, ulcerative colitis, functional heartburn, chronic intestinal pseudo-obstruction (or colonic pseudo-obstruction), visceral pain, abdominal pain, pelvic pain, anal fissure pain, pain associated with vulvodynia, pain associated with endometriosis, pain associated with fibromyalgia, functional abdominal pain, interstitial cystitis pain, diverticulitis, pain associated with diverticulitis, and pain associated with celiac sprue. In some embodiments, methods are provided to treat IBS-c, IBS-m or chronic constipation (e.g., chronic idiopathic constipation) in pediatric patients with the compositions and oral dosage forms described herein. In some

embodiments, methods are provided to treat IBS-c in a pediatric patient in need thereof. In some embodiments, methods are provided to treat chronic idiopathic constipation in a pediatric patient in need thereof.

[00080] In some embodiments, a method is for treating or relieving pain comprising administering to a patient in need thereof, a therapeutically effective amount of the composition described herein. In some embodiments, the pain is selected from visceral pain; abdominal pain; pelvic pain; or pain associated with gastrointestinal disorders, venereal diseases, bladder pain syndrome, diverticulitis pain, prostatitis, testicular pain, endometriosis, vulvodynia, rectal pain, or interstitial cystitis. In some embodiments, the pain is selected from pelvic pain, pain associated with proctitis, anal fissure pain, pain associated with vulvodynia, pain associated with endometriosis, pain associated with fibromyalgia, functional abdominal pain, interstitial cystitis pain, pain associated with venereal disease, diverticulitis, pain associated with diverticulitis, and pain associated with celiac sprue.

[00081]

[00082] In some embodiments, the effective dose range of linaclotide for adult humans is from 25 µg to 6 mg per day orally. In some embodiments, the dose range is 15 µg to 2 mg per day orally. In some embodiments, the dose range for adult humans is 15 µg to 1 mg per day orally (e.g., 15 µg, 30 µg, 50 µg, 72 µg, 100 µg, 145 µg, 150 µg, 200 µg, 250 µg, 290 µg, 300 µg, 350 µg, 400 µg, 450 µg, 500 µg, 550 µg, 579 µg, 600 µg, 650 µg, 700 µg, 750 µg, 800 µg, 850 µg, 900 µg, 950 µg or 1 mg). In some embodiments, the dose range for adult humans is 30 µg to 300 µg per day orally. In some embodiments, the dose range is 100 µg to 600 µg per day orally. In some embodiments, the dose is 30 µg, 100 µg, 150 µg, 200 µg, 300 µg, 400 µg, 500 µg or 600 µg linaclotide per day orally. In some embodiments, the dose is 50 µg linaclotide per day orally. In some embodiments, the dose is 100 µg linaclotide per day orally. In some embodiments, the dose is 145 µg linaclotide per day orally. In some embodiments, the dose is 200 µg linaclotide per day orally. In some embodiments, the dose is 290 µg linaclotide per day orally. In some embodiments, the dose is 300 µg linaclotide per day orally. In some embodiments, the dose is 500 µg linaclotide per day orally. In some embodiments, the dose is 600 µg linaclotide per day orally.

[00083] In some embodiments, the effective pediatric dose range of linaclotide is from 0.05 µg to 2 mg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.05 µg to 100 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to 90 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to 50 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to

25 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to 10 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to 5 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to 1 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is from 0.1 µg to 0.5 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 0.1 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 0.15 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 0.25 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 0.5 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 3.5 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 15 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 36 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 45 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 60 µg per day orally. In some embodiments, the effective pediatric dose range of linaclotide is 90 µg per day orally. In some embodiments, the unit dosage form and daily dose are equivalent.

[00084] In some embodiments, the unit dosage form is administered with food at any time of the day, without food at any time of the day, with food after an overnight fast (*e.g.*, with breakfast). In some embodiments, the unit dosage form is administered once a day, twice a day or three times a day. In some embodiments, one, two or three unit dosage forms will contain the daily oral dose of linaclotide. The precise amount of compound administered to a patient will be the responsibility of the attendant physician. However, the dose employed will depend on a number of factors, including the age and sex of the patient, the precise disorder being treated, and its severity.

[00085] In some embodiments, the compositions are administered as a monotherapy. In some embodiments, the composition consists essentially of an effective amount of linaclotide. In some embodiments, the composition consists of an effective amount of linaclotide.

[00086] In some embodiments, the compositions are directly administered to a patient, for example, in the form of delayed release tablet or delayed release capsule. In some embodiments, the compositions are dissolved, disintegrated and/or mixed on or within food or beverage prior to administration to patients (*e.g.*, elderly or pediatric patients). In some embodiments, the composition is dissolved or disintegrated in a liquid, solution, or fluid optionally containing stabilizing agent(s), preservative(s), sweetener(s), or the like, etc. prior to administration to a patient (*e.g.*, elderly or pediatric patient). In some embodiments, the composition is a multiple dose composition, *i.e.*, containing two, three, five, seven, ten, fifteen,

twenty, twenty-five, thirty, forty, fifty, sixty, seventy, eighty, ninety or more daily doses of linaclotide.

[00087] In other embodiments, the compositions are administered as part of a combination therapy. For example, a composition may be used in combination with other drugs or therapies that are used in the treatment, prevention, suppression, and/or amelioration of the diseases or conditions for which compounds of the invention are useful. The linaclotide can be co-administered or co-formulated with other medications. In one embodiment, the linaclotide composition can be co-administered with other medications used to treat gastrointestinal disorders including but not limited to acid suppressing agents such as Histamine-2 receptor agonists (H2As) and/or proton pump inhibitors (PPIs). In one embodiment, the linaclotide composition can be co-administered with other medications used to treat gastrointestinal disorders including 5-ASAs such as mesalamine.

[00088] Such other drug(s) may be administered, by a route and in an amount commonly used therefore, contemporaneously or sequentially with a compound of the invention. When a compound of the present invention is used contemporaneously with one or more other drugs, a pharmaceutical unit dosage form containing such other drugs in addition to the compound of the invention may be employed. Accordingly, the pharmaceutical compositions of the present invention include those that also contain one or more other active components, in addition to a compound of invention.

[00089] Several methods can be used for evaluating the bioactivity of the linaclotide composition, including, but not limited to, immunoassays (*e.g.*, enzyme-linked immunosorbent assay), radioimmuno assays, immunoradiometric assays, gel electrophoresis (*e.g.*, SDS-PAGE), high performance liquid chromatography (HPLC), and/or high performance capillary electrophoresis (HPCE). In some embodiments, the bioactivity of the composition is assessed by a method comprising fixing linaclotide, incubating linaclotide with guanylate cyclase C (GCC), incubating GCC bound linaclotide with antibodies against GCC, incubating GCC antibody-bound linaclotide with fluorescently labeled antibodies against GCC antibodies, and detecting the linaclotide bound to the GCC antibodies by measuring the fluorescence intensity using a plate reader. The drug concentration can then be calculated based on the fluorescence reading of the solution.

[00090] For example, the bioactivity of the linaclotide compositions can be assessed and quantified using the following method, though other methods are available. The composition is added to a volumetric flask containing 60 ml of phosphate buffer having a pH of 4.5, and the flask is shaken for 60 minutes. 0.2 ml of the supernatant is then removed, and is added into one or more wells of a 96-well plate that is coated with GC-C receptors. The plate is sealed and

incubated at 37°C for 2 hr. At the end of incubation, the sample is removed and the plate is washed with phosphate buffered saline (PBS). The bound linacotide is then incubated for 1 hour, at room temperature, with GC-C (such as is available from Sigma-Aldrich Inc.) labeled with fluorescein isocyanate (FITC) in blocking buffer. After incubation, the well is washed with PBS. The fluorescence intensity of the end product is detected, for example, by using a plate reader. The linacotide concentration is then calculated based on the fluorescence reading of the solution.

[00091] *B. Delayed Release Compositions*

[00092] Delayed release oral dosage forms of linacotide (collectively, “DR”) are provided herein. The delayed release pharmaceutical compositions of the present invention relates to stable, solid, oral dosage forms of linacotide which exhibit delayed release of linacotide to the lower gastrointestinal tract. Until now, the only approved formulation of linacotide is a capsule that exhibits immediate release (“IR”). These IR dosage forms release most or all of the linacotide contained therein in the upper GI. This, in turn, causes GC-C receptor activation and fluid secretion in both the upper GI and to a lesser extent in the lower GI. The difference between upper and lower GI activation and fluid secretion by the IR dosage form is due, in part, to the fact that linacotide (once released from the dosage form) undergoes proteolytic digestion and loses some or all capacity to activate GC-C receptors, particularly by the time it reaches the lower GI (such as the ileum, terminal ileum, ileocecal valve, or colon).

[00093] In some embodiments, the linacotide is present in the composition in an amount between 30 µg to 1,000 µg. For example, in some embodiments, the linacotide is present in an amount of about 30 µg, about 100 µg, about 300 µg, about 500 µg, about 600 µg, or about 1,000 µg or the linacotide is present in an amount of 100 µg, 300 µg, 500 µg, 600 µg, or 1,000 µg.

[00094] In some embodiments, the composition further comprises between 0% - 2% per weight of an amino acid selected from the group consisting of alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, and valine, or any mixture thereof. In some embodiments, the composition further comprises between 0% - 2% or between 0.5% - 1.5% per weight of histidine. In some embodiments, the composition further comprises between 0% - 3% per weight of a cation salt selected from the group consisting of calcium, potassium, magnesium, zinc, aluminum, manganese, chromium, cobalt, nickel, barium, and sodium, or any combination or mixture thereof. In some embodiments, the composition further comprises between 0% - 3% per weight of a calcium salt. In some

embodiments, the composition further comprises between 0% - 2% or between 0.2% - 0.8% per weight of calcium chloride dehydrate. In some embodiments, the composition further comprises between 0% - 5%, between 1% - 3%, or between 1% - 1.88% per weight of polyvinyl alcohol (PVA).

[00095] In some embodiments, the pH-sensitive polymer has a dissolution pH of at least 6.0, at least 6.5, or at least 7.0. In some embodiments, The the pH-sensitive polymer comprises methyl acrylate-methacrylic acid copolymers (e.g., Eudragit®). In some embodiments, the pH-sensitive polymer comprises Eudragit S100. In some embodiments, the pH-sensitive polymer comprises Eudragit L100. In some embodiments, the pH-sensitive polymer consists essentially of Eudragit S100. In some embodiments, the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100. In some embodiments, the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100 at a ratio of between 1:1 and 6:1 (S100:L100), at a ratio of between 4.5:1 and 5.5:1 (S100:L100), or at a ratio of 4.875:1 (S100:L100) by weight.

[00096] In some embodiments, the delayed release pharmaceutical tablet composition comprises an enteric coated tablet. In some embodiments, the delayed release pharmaceutical tablet composition comprises:

Ca²⁺;
histidine; and
polyvinyl alcohol (PVA).

[00097] In some embodiments, the composition further comprises a protective polymer film or subcoating. In some embodiments, the subcoating comprises Opadry II®.

[00098] The DR dosage forms described herein release most or all of the linaclotide contained therein within the lower GI, such as proximate to the ileocecal valve or within the colon (and less or no release in the stomach, duodenum and/or jejunum). Therefore, the inventive dosage forms have a capacity to achieve lower overall fluid secretion than IR dosage forms in the upper GI, while improving or still maintaining excellent efficacy for treating a disorder (e.g., a GI disorder such as IBS-c or symptoms associated with a disorder, such as abdominal pain). IBS patients report lower left quadrant abdominal pain as a symptom of their disorder, so it is believed that the pain of IBS originates from the colon. Moreover, the DR dosage forms are believed to be ideally suited for treating lower GI-associated diseases and disorders. Because the DR dosage forms will not release any (or a small percentage) of its linaclotide in the stomach and upper GI (which can cause rapid digestion of the linaclotide in the intestine), some preferred embodiments of the DR dosage form will incorporate low doses

of linaclotide (as compared to the amounts in the approved IR form) but will maintain the same efficacy levels as the IR in treating GI symptoms. Disorders that are suitable for treatment with the delayed release compositions include irritable bowel syndrome (IBS), constipation, irritable bowel syndrome with constipation (IBS-c), irritable bowel syndrome with diarrhea (IBS-d), mixed IBS (IBS-m), un-subtyped IBS (IBS-u), chronic idiopathic constipation (CIC), diverticulitis, ulcerative colitis, a functional gastrointestinal disorder, gastroesophageal reflux disease, functional heartburn, dyspepsia, visceral pain, abdominal pain, gastroparesis, chronic intestinal pseudo-obstruction, colonic pseudo-obstruction, Crohn's disease, inflammatory bowel disease, overactive bladder syndrome, bladder hypersensitivity or colitis induced bladder afferent hyperactivity in a patient in need thereof. Symptoms that are suitable for treatment with the delayed release compositions include abdominal pain, discomfort or bloating, or visceral pain, for example, in a non-constipated patient.

[00099] In general, the guanylate cyclase C (GC-C) receptor is a transmembrane receptor that is located on the apical surface of epithelial cells in the stomach, intestine and lower GI. The receptor has an extracellular ligand-binding domain, a single transmembrane region and a C-terminal guanylyl cyclase domain. When a ligand binds to the extracellular domain of GC-C, the intracellular catalytic domain catalyzes the production of cGMP from GTP. In vivo, this increase in intracellular cGMP initiates a cascade of events that leads to, among other things, increased secretion of chloride and bicarbonate into the intestinal lumen, increased luminal pH, decreased luminal sodium absorption, increased fluid secretion, and acceleration of intestinal transit. cGMP is secreted bi-directionally from the epithelium into the submucosa and lumen. Normally, the pH of the GI tract gradually increases from stomach (pH 1.5-3) to terminal ileum (pH 7-8) before it drops in the colon to pH 5.5-7.0. In addition, there is growing evidence that the potent analgesic effects of linaclotide *in vivo* are mediated by a pathway linking extracellular cGMP, secreted from IEC (intestinal epithelial cells) into the submucosa following activation of the GC-C/cGMP pathway by linaclotide, to altered function of colonic nociceptors resulting in peripheral analgesia.

[000100] Linaclotide binds to the intestinal GC-C receptor which is a regulator of fluid and electrolyte balance in the intestine. Linaclotide is a peptide that consists of the amino acid sequence Cys₁ Cys₂ Glu₃ Tyr₄ Cys₅ Cys₆ Asn₇ Pro₈ Ala₉ Cys₁₀ Thr₁₁ Gly₁₂ Cys₁₃ Tyr₁₄. Any desired form of linaclotide may be used in the composition, for example, any pharmaceutically acceptable salt or hydrate of the peptide, any isolated and/or purified form thereof, or any disulfide form thereof. Linaclotide has disulfide bonds between Cys₁ and Cys₆, Cys₂ and Cys₁₀, and Cys₅ and Cys₁₃.

[000101] In some embodiments, the DR composition comprises enteric-coated tablets comprising an immediate release tablet core and containing a unit dose of linaclotide that dissolves only under pH conditions of the distal segment of intestine. In some embodiments, the enteric or functional coating comprises a pH-sensitive polymer.

[000102] The pH-sensitive polymer is chosen on the basis of the threshold pH (or dissolution pH) consistent with the pH of the part of the GI tract where release is desired. Therefore, in one embodiment, the enteric coating comprises a pH-sensitive polymer that has a dissolution profile of a pH of at least 6.0, for example, a pH of at least 6.2, a pH of at least 6.4, a pH of at least 6.5, a pH of at least 6.6, a pH of at least 6.8, a pH of at least 7.0, a pH of at least 7.2, a pH of at least 7.4, a pH of at least 7.6 or higher.

[000103] In another embodiment, the pH-sensitive polymer is selected from methyl acrylate-methacrylic acid copolymers (e.g. Eudragit®); cellulose acetate succinate (CAS); hydroxy propyl methyl cellulose phthalate (HPMCP); PVA; PVP; PVP-LP, hydroxy propyl methyl cellulose acetate succinate (HPMCAS); polyvinyl acetate phthalate (PVAP); methyl methacrylate-methacrylic acid copolymers; sodium alginate and stearic acid; guar gum; and carbomers. In further embodiments, the enteric coating is selected from Eudragit® FS30D, PlasAcryl®, Eudragit® S100, Eudragit®L100, Eudragit®L100-55, Eudragit® L30D-55, Eudragit® S, Eudragit®RL30D, Eudragit®RS30D, Eudragit® RS, Eudragit® EC, or mixture thereof. In one embodiment, the pH-sensitive polymer comprises Eudragit S100. In another embodiment, the pH-sensitive polymer comprises Eudragit L100. In still another embodiment, the pH-sensitive polymer consists essentially of Eudragit S100. In still another embodiment, the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100. In still another embodiment, the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100 at a ratio of between 1:1 and 6:1 (S100:L100) by weight. In another embodiment, the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100 at a ratio of between 4.5:1 and 5.5:1 (S100:L100) by weight. In one particular embodiment, the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100 at a ratio of 4.875:1 (S100:L100) by weight.

[000104] In yet another embodiment, the enteric coating is at least 40 microns in average thickness, for example, at least 45 microns in average thickness, at least 50 microns in average thickness, at least 55 microns in average thickness, at least 60 microns in average thickness, at least 65 microns in average thickness, at least 70 microns in average thickness, at least 75 microns in average thickness, at least 80 microns in average thickness, at least 85 microns in average thickness, at least 90 microns in average thickness, at least 95 microns in average thickness, at least 100 microns in average thickness, at least 105 microns in average thickness,

at least 110 microns in average thickness, at least 115 microns in average thickness, or at least 120 microns in average thickness. In another embodiment, the enteric coating has an average thickness of between 55 microns and 100 microns. In still another embodiment, the enteric coating has an average thickness of between 65 microns and 95 microns. In a particular embodiment, the enteric coating has an average thickness of about 75 microns and 85 microns.

[000105] In some embodiments, the delayed release composition comprises at least 1.25% (w/w) of PVA, for example, at least 1.49% (w/w) of PVA. In some embodiments, the delayed release composition comprises at least 0.44% (w/w) of CaCl_2 , for example, at least 0.71% (w/w) of CaCl_2 . In some embodiments, the delayed release composition comprises at least 0.93% (w/w) of histidine, for example, at least 1.49% (w/w) of histidine.

[000106] The delayed release compositions may include any effective amount of linaclotide. In some embodiments, for example, the composition comprises from 0.05 μg to 6 mg of linaclotide. In some embodiments, for example, the composition comprises from 1 μg to 2 mg of linaclotide. In some embodiments, the composition comprises from 25 μg to 2 mg of linaclotide, for example, from 50 μg to 1 mg of linaclotide. In some embodiments, for example, the composition comprises from 0.1 μg to 90 μg of linaclotide. In some embodiments, for example, the composition comprises from 0.1 μg to 45 μg of linaclotide. In some embodiments, for example, the composition comprises from 0.1 μg to 25 μg of linaclotide. In some embodiments, for example, the composition comprises from 30 μg to 300 μg of linaclotide. In some embodiments, the composition comprises 0.05 μg , 0.1 μg , 0.15 μg , 0.25 μg , 0.5 μg , 0.75 μg , 1 μg , 1.5 μg , 2 μg , 2.5 μg , 3 μg , 3.5 μg , 4 μg , 4.5 μg , 5 μg , 7.5 μg , 9 μg , 10 μg , 15 μg , 20 μg , 25 μg , 30 μg , 35 μg , 36 μg , 40 μg , 45 μg , 50 μg , 60 μg , 72 μg , 75 μg , 90 μg , 100 μg , 145 μg , 150 μg , 200 μg , 250 μg , 290 μg , 300 μg , 350 μg , 400 μg , 450 μg , 500 μg , 550 μg , 579 μg , 600 μg , 650 μg , 700 μg , 750 μg , 800 μg , 850 μg , 900 μg , 950 μg or 1 mg of linaclotide. In some embodiments, the composition comprises from 100 μg to 600 μg of linaclotide. In some embodiments, the composition comprises 30 μg , 50 μg , 75 μg , 100 μg , 150 μg , 290 μg , 300 μg , 400 μg , 500 μg or 600 μg of linaclotide. In some embodiments, the composition comprises 9 μg , 10 μg , 15 μg , 36 μg , 72 μg , 75 μg , 145 μg , 290 μg , 579 μg , or 600 μg of linaclotide.

[000107] In some embodiments, the composition comprises 30 μg of linaclotide. In some embodiments, the composition comprises 50 μg of linaclotide. In some embodiments, the composition comprises 100 μg of linaclotide. In some embodiments, the composition comprises 145 μg of linaclotide. In some embodiments, the composition comprises 300 μg of linaclotide.

[000108] It has been found, in some embodiments, that the stability of delayed release compositions of linacotide can be increased or improved by including in the compositions a suitable amount of a sterically hindered primary amine (*e.g.*, amino acid) component, a cation (*e.g.*, metal cation) component, and/or a polymer component. These components increase or enhance the stability of delayed release compositions of linacotide, for example, by preventing, lessening, and/or decreasing degradation of linacotide within the composition (for example, due to moisture-driven degradation reactions, *e.g.*, hydrolysis, deamidation, and/or multimerization reactions). For instance, it has been found in some embodiments that addition or inclusion of a suitable amount of a cation (*e.g.*, Mg^{2+} , Ca^{2+} , Zn^{2+}) in the composition increases the stability of the composition against oxidative degradation of linacotide. Moreover, it has been found in some embodiments that inclusion of a suitable amount of a sterically hindered primary amine for an example in the form of an amino acid (*e.g.*, histidine) in the composition increases the stability of the composition against, for example, the nucleophilic addition of formaldehyde or a formaldehyde equivalent to the N-terminus of linacotide, *e.g.* by acting as a scavenger, and/or by buffering the composition. Moreover, it has been found in some embodiments that inclusion of both a sterically hindered primary amine (*e.g.*, histidine) and a cation (*e.g.*, Ca^{2+}) in suitable amounts in the composition increases the stability of the composition against the formation of hydrolysis and formaldehyde (Cys¹-IMD) products of linacotide. It has also been found in some embodiments that inclusion of a suitable amount of a polymer (*e.g.*, polyvinyl pyrrolidone or polyvinyl alcohol) in the delayed release composition increases the stability of the composition for example by decreasing the mobility and/or reactivity of linacotide within the composition, *e.g.*, by forming a complex or matrix (for example, a glassy and/or rigid matrix) with linacotide (*e.g.*, by vitrification reaction), by preventing or lessening hydrogen bond formation between linacotide and water molecules, and/or by enhancing the three-dimensional structural integrity of linacotide.

[000109] In this regard, it has been found in some embodiments that combining linacotide in an delayed release pharmaceutical composition with specific concentrations or molar ratios of the cation and sterically hindered primary amine causes a synergistic enhancement or improvement in the stability of linacotide within the composition, for example as compared to similar compositions not containing the cation and/or sterically hindered primary amine and/or the same concentrations of these components. In some embodiments, composition can comprise any stabilizing amount of a sterically hindered primary amine component. In other embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 400:1 and 1:1. In further, embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide

between 200:1 and 50:1. In other embodiments, the composition can comprise a molar ratio of sterically hindered primary amine (*e.g.*, amino acid) to linacotide between 100:1 and 1:100. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 1:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 90:1 and 2:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 80:1 and 5:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 70:1 and 10:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 60:1 and 20:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 50:1 and 30:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 40:1 and 20:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 20:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 25:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 30:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 40:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 50:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 60:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide between 100:1 and 70:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide of at least 5:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide of at least 10:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide of at least 20:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide of at least 25:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide of at least 30:1. In some embodiments, the composition comprises a molar ratio of sterically hindered primary amine to linacotide of at least 40:1.

[000110] Suitable sterically hindered primary amines for inclusion in the delayed release composition are, for example, naturally-occurring amino acids (*e.g.*, alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine,

leucine, lysine, meglumine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine), synthetic amino acids (*e.g.*, lanthionine, theanine or 1-amino cyclohexane), amino sugars (*e.g.*, chitosan or glucosamine), or combination or mixtures thereof. In some embodiments, the composition comprises an amino acid selected from alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine, or a mixture thereof. In some embodiments, the composition comprises an amino acid selected from leucine, isoleucine, asparagine, glutamine, glutamic acid, histidine, cysteine, alanine, serine, threonine, tyrosine, proline, tryptophan, or a combination or mixture thereof. In some embodiments, the composition comprises an amino acid selected from leucine, isoleucine, methionine, alanine, or a combination or mixture thereof. In some embodiments, the composition comprises an amino acid selected from leucine, isoleucine, alanine, or a combination or mixture thereof. In some embodiments, the composition comprises an amino acid selected from leucine, isoleucine, methionine, or a combination or mixture thereof. In some embodiments, the composition comprises an amino acid selected from leucine, methionine, alanine, or a combination or mixture thereof. In some embodiments, the composition comprises leucine, methionine, or a mixture thereof. In some embodiments, the composition comprises leucine, isoleucine, or a mixture thereof. In some embodiments, the composition comprises leucine, alanine, or a mixture thereof. In some embodiments, the composition comprises leucine. In some embodiments, the composition comprises isoleucine. In some embodiments, the composition comprises methionine. In some embodiments, the composition comprises alanine. In some embodiments, the composition comprises histidine.

[000111] The delayed release composition can comprise any stabilizing amount of a cation (*e.g.*, metal cation). In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 300:1 and 1:1. In further embodiments, the composition comprises a molar ratio of cation to linaclotide between 250:1 and 30:1. In other embodiments, the composition can comprise a molar ratio of cation to linaclotide between 100:1 and 1:100. In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 100:1 and 1:1. In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 90:1 and 2:1. In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 80:1 and 5:1. In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 70:1 and 10:1. In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 60:1 and 20:1. In some embodiments, the composition comprises a molar ratio of cation to linaclotide between 50:1 and 30:1. In some embodiments, the composition comprises a molar ratio of cation to

linacotide between 40:1 and 20:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 20:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 25:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 30:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 40:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 50:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 60:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide between 100:1 and 70:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 5:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 10:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 20:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 25:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 30:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 40:1. In some embodiments, the composition comprises a molar ratio of cation to linacotide of at least 60:1.

[000112] Any suitable cation(s) can be included in the composition, for example, any suitable metal cation or organic cation. In some embodiments, the composition comprises a metal cation selected from calcium, potassium, magnesium, zinc, aluminum, iron, tin, manganese, chromium, cobalt, nickel, barium, sodium, or a combination or mixture thereof. In some embodiments, the composition comprises a metal cation selected from calcium, potassium, magnesium, zinc, aluminum, manganese, chromium, cobalt, nickel, barium, sodium, or a combination or mixture thereof. In some embodiments, the composition comprises a metal cation selected from aluminum, calcium, potassium, sodium, magnesium, manganese, zinc, or a combination or mixture thereof. In some embodiments, the composition comprises a metal cation selected from calcium, magnesium, manganese, zinc, or a combination or mixture thereof. In some embodiments, the composition comprises a divalent metal cation. In some embodiments, the composition comprises a divalent metal cation selected from Al^{3+} , Ca^{2+} , Mg^{2+} , Zn^{2+} , Mn^{2+} , or a combination or mixture thereof. In some embodiments, the composition comprises Mg^{2+} . In some embodiments, the composition comprises Ca^{2+} . In some embodiments, the composition comprises Zn^{2+} . In some embodiments, the composition comprises aluminum.

[000113] Moreover, the metal cation can be added to the composition in any suitable form, for example any pharmaceutically acceptable salt with any appropriate counterion.

Suitable metal salts include, for example, calcium chloride, calcium carbonate, calcium acetate, magnesium chloride, magnesium acetate, zinc acetate, zinc chloride, or mixtures thereof. In some embodiments, the composition comprises calcium chloride, magnesium chloride, zinc acetate, or any combination or mixture thereof. In some embodiments, the composition comprises calcium chloride. In some embodiments, the composition comprises magnesium chloride. In some embodiments, the composition comprises zinc acetate. Suitable organic cations include, for example, ammonium hydroxide, D-arginine, L-arginine, t-butylamine, calcium acetate hydrate, calcium carbonate, calcium DL-malate, calcium hydroxide, choline, ethanolamine, ethylenediamine, glycine, L-histidine, L-lysine, magnesium hydroxide, N-methyl-D-glucamine, L-ornithine hydrochloride, potassium hydroxide, procaine hydrochloride, L-proline, pyridoxine, L-serine, sodium hydroxide, DL-tryptophan, tromethamine, L-tyrosine, L-valine, carnitine, taurine, creatine malate, arginine alpha ketoglutarate, ornithine alpha ketoglutarate, spermine acetate, spermidine chloride, or combinations or mixtures thereof. In some embodiments, the organic cation is selected from the group consisting of N-methyl D-glucamine, choline, arginine, lysine, procaine, tromethamine (TRIS), spermine, N-methyl-morpholine, glucosamine, N,N-bis(2-hydroxyethyl) glycine, diazabicycloundecene, creatine, arginine ethyl ester, amantadine, rimantadine, ornithine, taurine, and citrulline, or any combination or mixture thereof.

[000114] The composition can contain any stabilizing amount of a polymer. In some embodiments, the composition comprises between 1 and 25 % by weight of a polymer, relative to the total weight of the composition. In some embodiments, the composition comprises between 1 and 10% by weight of a polymer, relative to the total weight of the composition.

[000115] In some embodiments, the composition comprises between 2 and 4 % by weight of a polymer, relative to the total weight of the composition. In some embodiments, the composition comprises between 0.1 and 75 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 55 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 35 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 30 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 25 wt.% of a polymer. In some embodiments, the composition comprises between 1 and 25 wt.% of a polymer. In some embodiments, the composition comprises between 5 and 25 wt.% of a polymer. In some embodiments, the composition comprises between 10 and 25 wt.% of a polymer. In some embodiments, the composition comprises between 15 and 25 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 22 wt.% of a polymer. In some embodiments, the composition comprises between 1 and 22 wt.% of a polymer. In some embodiments, the

composition comprises between 5 and 22 wt.% of a polymer. In some embodiments, the composition comprises between 10 and 22 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 20 wt.% of a polymer. In some embodiments, the composition comprises between 1 and 20 wt.% of a polymer. In some embodiments, the composition comprises between 5 and 20 wt.% of a polymer. In some embodiments, the composition comprises between 10 and 20 wt.% of a polymer. In some embodiments, the composition comprises between 0.01 and 15 wt.% of a polymer. In some embodiments, the composition comprises between 0.01 and 10 wt.% of a polymer. In some embodiments, the composition comprises between 0.01 and 5 wt.% of a polymer. In some embodiments, the composition comprises between 0.1 and 95 wt.%, for example, between 5 and 95 wt.%, between 15 and 95 wt.%, between 25 and 95 wt.%, between 35 and 95 wt.%, between 45 and 95 wt.%, between 0.1 and 85 wt.%, between 1 and 85 wt.%, between 5 and 85 wt.%, between 15 and 85 wt.%, between 25 and 85 wt.%, between 35 and 85 wt.%, between 0.1 and 80 wt.%, between 1 and 80 wt.%, between 5 and 80 wt.%, between 15 and 80 wt.%, between 25 and 80 wt.%, between 35 and 80 wt.%, between 0.1 and 75 wt.%, between 1 and 75 wt.%, between 5 and 75 wt.%, between 15 and 75 wt.%, between 25 and 75 wt.%, between 35 and 75 wt.%, between 0.1 and 65 wt.%, between 1 and 65 wt.%, between 5 and 65 wt.%, between 15 and 65 wt.%, between 25 and 65 wt.%, between 35 and 65 wt.%, between 0.1 and 60 wt.%, between 1 and 60 wt.%, between 5 and 60 wt.%, between 15 and 60 wt.%, between 25 and 60 wt.%, or between 35 and 60 wt.% of a polymer.

[000116] In some embodiments, the polymer acts as both a stabilizer, protective coating, or as a film forming agent within the delayed release composition. In some embodiments, the delayed release composition comprises a molar ratio of polymer (e.g., PVP or PVA) to linacotide between 80:1 and 300:1, for example, between 100:1 and 200:1, between 110:1 and 190:1, or even between 120:1 and 180:1. In some embodiments, the delayed release composition comprises a molar ratio of polymer (e.g., PVP or PVA) to linacotide greater than about 80:1, for example, greater than about 100:1, or even greater than about 120:1. In some embodiments, the delayed release composition comprises a weight ration of polymer (e.g., PVP or PVA) to linacotide between 10:1 and 300:1, for example, between 80:1 and 200:1, between 100:1 and 180:1, or even between 110:1 and 150:1. In some embodiments, the delayed release composition comprises a weight ration of polymer (e.g., PVP or PVA) to linacotide between 100:1 and 500:1, for example, between 200:1 and 400:1, between 250:1 and 350:1, or even between 300:1 and 350:1.

[000117] Suitable polymers for inclusion in the delayed release compositions are, for example, polyvinyl pyrrolidone (PVP), polyvinyl alcohol (PVA), polyvinyl alcohol low

peroxide (PVA-LP), hydroxylpropyl methyl cellulose (HPMC), hydroxylpropyl cellulose (HPC), methyl cellulose, methacrylate polymers, cyclodextrin, dextrin, dextran, polyacrylic acid, chitosan, guar gum, xanthan gum, polyethylene oxide (*e.g.*, polyethylene polypropylene oxide), poly (sodium vinylsulfonate), polyethylene glycol, poly(arginine), poly carbophil, polyvinyl pyrrolidone-co-vinyl acetate, a poloxamer (*e.g.*, Pluronic® products available from BASF), alginate, trehalose, sucrose, inulin, or a combination or mixture thereof. In some embodiments, the composition comprises a polymer selected from PVP, PVA, methacrylate polymers, cyclodextrin, dextran, polyacrylic acid, chitosan, guar gum, xanthan gum, polyethylene oxide, polyethylene glycol, poly(arginine), poly carbophil, polyvinyl pyrrolidone-co-vinyl acetate, a poloxamer, or a combination or mixture thereof. In some embodiments, the composition comprises PVP, PVA, polyethylene oxide, or a mixture thereof. In some embodiments, the composition comprises PVP, PVA, or a mixture thereof. In some embodiments, the composition comprises PVP. In some embodiments, the composition comprises PVA.

[000118] In some embodiments, the delayed release composition comprises two or more stabilizing agents. For example, the composition can include a stabilizing amount of a polymer and a stabilizing amount of a sterically hindered primary amine. Moreover, the composition can include a stabilizing amount of a polymer and a stabilizing amount of a cation (*e.g.*, metal cation). In addition, the composition can include a stabilizing amount of a sterically hindered primary amine and a stabilizing amount of a cation (*e.g.*, metal cation). In some embodiments, the composition comprises a stabilizing amount of a polymer, a stabilizing amount of a sterically hindered primary amine, and a stabilizing amount of a cation (*e.g.*, metal cation).

[000119] In some embodiments, the delayed release composition comprises a stabilizing amount of PVP and a stabilizing amount of an amino acid selected from histidine, alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine, or a mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of an amino acid selected from alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine, or a mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of leucine, isoleucine, methionine, alanine, or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of histidine.

[000120] In some embodiments, the delayed release composition comprises a stabilizing amount of PVA and a stabilizing amount of an amino acid selected from alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, glycine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine, or a mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of an amino acid selected from alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, valine, or a mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of leucine, isoleucine, methionine, alanine, or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of leucine.

[000121] In some embodiments, the delayed release composition comprises a stabilizing amount of PVP and a stabilizing amount of a cation (*e.g.*, metal cation). In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of a divalent metal cation. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of Mg^{2+} , Ca^{2+} , Zn^{2+} or a salt thereof or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of Ca^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of Mg^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVP and a stabilizing amount of Zn^{2+} or a salt thereof.

[000122] In some embodiments, the delayed release composition comprises a stabilizing amount of PVA and a stabilizing amount of a cation (*e.g.*, metal cation). In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of a divalent metal cation. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of Mg^{2+} , Ca^{2+} , Zn^{2+} or a salt thereof or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of Ca^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of Mg^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVA and a stabilizing amount of Zn^{2+} or a salt thereof.

[000123] In some embodiments, the delayed release composition comprises a stabilizing amount of an amino acid selected from leucine, isoleucine, methionine, alanine; and a stabilizing amount of a divalent metal cation selected from Mg^{2+} , Ca^{2+} , Zn^{2+} or a salt thereof

or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of an amino acid selected from leucine, and isoleucine; and a stabilizing amount of a divalent metal cation selected from Mg^{2+} , Ca^{2+} or a salt thereof or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of an amino acid selected from leucine or methionine; and a stabilizing amount of a divalent metal cation selected from Ca^{2+} , Zn^{2+} or a salt thereof or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of leucine and a stabilizing amount of Ca^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of a cation and a stabilizing amount of a sterically hindered primary amine. In some embodiments, the composition comprises a cation and a sterically hindered primary amine in a molar ratio of cation:sterically hindered primary amine (e.g., Ca^{2+} :leucine) of at least 1.5:1, e.g., at least 2:1, at least 2.5:1, at least 3:1, at least 4:1, or even at least 5:1 (for example, a molar ratio between 1.5:1 and 5:1, e.g., between 2:1 and 4:1).

[000124] In some embodiments, the delayed release composition comprises (i) a stabilizing amount of PVP or PVA, (ii) a stabilizing amount of leucine, isoleucine, methionine, alanine, and (iii) a stabilizing amount of Mg^{2+} , Ca^{2+} , Zn^{2+} or a salt thereof or a combination or mixture thereof. In some embodiments, the composition comprises a stabilizing amount of PVP, a stabilizing amount of leucine, and a stabilizing amount of a metal cation. In some embodiments, the composition comprises a stabilizing amount of PVA, a stabilizing amount of histidine, and a stabilizing amount of Ca^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVP, a stabilizing amount of leucine, and a stabilizing amount of Mg^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVP, a stabilizing amount of leucine, and a stabilizing amount of Zn^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVA, a stabilizing amount of leucine, and a stabilizing amount of Ca^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVA, a stabilizing amount of leucine, and a stabilizing amount of Mg^{2+} or a salt thereof. In some embodiments, the composition comprises a stabilizing amount of PVA, a stabilizing amount of leucine, and a stabilizing amount of Zn^{2+} or a salt thereof.

[000125] In some embodiments, the composition comprises (i) between 0.1 and 30 wt.% of a polymer, (ii) a sterically hindered primary amine (e.g., an amino acid) in a molar ratio of primary amine to linaclotide between 100:1 and 10:1, and (iii) a cation (e.g., a metal cation) in a molar ratio of cation to linaclotide between 100:1 and 40:1. In some embodiments, the composition comprises (i) between 5 and 25 wt.% of a polymer, (ii) a sterically hindered primary amine (e.g., an amino acid) in a molar ratio of primary amine to linaclotide 100:1 and

30:1 (*e.g.*, between 60:1 and 30:1 or even between 50:1 and 30:1), and (iii) a cation (*e.g.*, a metal cation) in a molar ratio of cation to linacotide between 100:1 and 60:1. In some embodiments, the composition comprises (i) between 0.1 and 30 wt.% of a polymer selected from PVP and PVA, (ii) an amino acid selected from leucine, isoleucine, alanine, and methionine in a molar ratio of amino acid to linacotide between 100:1 and 10:1, and (iii) a metal cation selected from Ca^{2+} , Mg^{2+} , and Zn^{2+} in a molar ratio of cation to linacotide between 100:1 and 40:1. In some embodiments, the composition comprises (i) between 5 and 25 wt.% of a polymer selected from PVP and PVA, (ii) an amino acid selected from leucine, isoleucine, alanine, and methionine in a molar ratio of amino acid to linacotide 100:1 and 30:1 (*e.g.*, between 60:1 and 30:1), and (iii) a metal cation selected from Ca^{2+} , Mg^{2+} , and Zn^{2+} in a molar ratio of cation to linacotide between 100:1 and 60:1. In some embodiments, the composition comprises (i) between 0.1 and 30 wt.% (*e.g.*, between 5 and 25 wt.%) of PVP or PVA, (ii) leucine in a molar ratio of leucine to linacotide between 100:1 and 30:1 (*e.g.*, between 60:1 and 30:1 or even between 50:1 and 30:1), and (iii) Ca^{2+} in a molar ratio of Ca^{2+} to linacotide between 100:1 and 60:1.

[000126] In some embodiments, the composition comprises (i) between 45 and 99 wt.% of a polymer, (ii) a sterically hindered primary amine (*e.g.*, an amino acid) in a molar ratio of primary amine to linacotide between 100:1 and 10:1, and (iii) a cation (*e.g.*, a metal cation) in a molar ratio of cation to linacotide between 100:1 and 40:1. In some embodiments, the composition comprises (i) between 45 and 70 wt.% of a polymer, (ii) a sterically hindered primary amine (*e.g.*, an amino acid) in a molar ratio of primary amine to linacotide 100:1 and 30:1 (*e.g.*, between 60:1 and 30:1 or even between 50:1 and 30:1), and (iii) a cation (*e.g.*, a metal cation) in a molar ratio of cation to linacotide between 100:1 and 60:1. In some embodiments, the composition comprises (i) between 45 and 99 wt.% of a polymer selected from PVP and PVA, (ii) an amino acid selected from leucine, isoleucine, alanine, and methionine in a molar ratio of amino acid to linacotide between 100:1 and 10:1, and (iii) a metal cation selected from Ca^{2+} , Mg^{2+} , and Zn^{2+} in a molar ratio of cation to linacotide between 100:1 and 40:1. In some embodiments, the composition comprises (i) between 45 and 70 wt.% of a polymer selected from PVP and PVA, (ii) an amino acid selected from leucine, isoleucine, alanine, and methionine in a molar ratio of amino acid to linacotide 100:1 and 30:1 (*e.g.*, between 60:1 and 30:1), and (iii) a metal cation selected from Ca^{2+} , Mg^{2+} , and Zn^{2+} in a molar ratio of cation to linacotide between 100:1 and 60:1. In some embodiments, the composition comprises (i) between 45 and 99 wt.% (*e.g.*, between 45 and 70 wt.%) of PVP or PVA, (ii) leucine in a molar ratio of leucine to linacotide between 100:1 and 30:1 (*e.g.*, between 60:1

and 30:1 or even between 50:1 and 30:1), and (iii) Ca^{2+} in a molar ratio of Ca^{2+} to linacotide between 100:1 and 60:1.

[000127] The delayed release composition (*e.g.*, delayed release tablet) may also comprise any one or more filling agents. Suitable filling agents include, but are not limited to, starch, calcium carbonate, calcium sulfate, hydroxypropylmethyl cellulose, fructose, methyl cellulose, dextrans, dextrose, dextran, lactitol, maltose, sucrose, sorbitol, isomalt, pregelatinized starch, dicalcium phosphate, microcrystalline cellulose, mannitol, gelatin, trehalose, erythritol, maltitol, lactose, glucose, or a combination thereof, or a mixture thereof. In some embodiments, the filling agent is isomalt. In some embodiments, the filling agent is gelatin. In some embodiments, the filling agent is mannitol. In some embodiments, the filling agent is pregelatinized starch. In some embodiments, the filling agent is microcrystalline cellulose.

[000128] The delayed release composition (*e.g.*, delayed release tablet) can comprise any suitable concentration of filling agent. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 0.1-99 % by weight, relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 1-95 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 10-90 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 20-90 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 25-85 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 30-80 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 40-70 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 10-60 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, for example, the composition comprises one or more filling agents in a concentration of 20-50 wt.% of filling agent(s), relative to the total weight of the composition. In some embodiments, the composition comprises one or more filling agents in a concentration of at least 20 wt.%, for example, at least 40 wt.%, at least 60 wt.%, at least 70 wt.%, at least 80 wt.%, or at least 90 wt.%, relative to the total weight of the composition.

[000129] In some embodiments, the delayed release composition (e.g., delayed release film) comprises one or more plasticizers. Suitable plasticizers include, but are not limited to, polyethylene glycol, propylene glycol, glycerin, glycerol, monoacetin, diacetin, triacetin, dimethyl phthalate, diethyl phthalate, dibutyl phthalate, dibutyl sebacate, triethyl citrate, tributyl citrate, triethyl citrate, triethyl acetyl citrate, castor oil, acetylated monoglycerides, sorbitol or combinations thereof. In exemplary embodiments, the concentration of the plasticizer in the formulation may be about 0 to about 30 wt.%, for example, about 1 to about 20 wt.%, about 0 to about 10 wt.%, about 1 to about 5 wt.%, or even 0 to about 4 wt.%.

[000130] In some embodiments, the delayed release composition comprises a film forming agent, a water-soluble polymer, a pH sensitive polymer, biodegradable polymer, or combination thereof. Water soluble, pH sensitive, or biodegradable polymers that may be used in the orally dissolving formulations of the present invention include, but are not limited to, cellulose derivatives, synthetic polymers polyacrylates and natural gums. For example, the water soluble polymers used in the orally dissolving formulations of the present invention may include, but are not limited to, methyl cellulose, hydroxypropyl cellulose, hydroxypropylmethyl cellulose, ethyl cellulose, hydroxyethyl cellulose, hydroxypropyl cellulose, carboxymethyl cellulose, cellulose acetate phthalate, cellulose acetate butyrate, amylose, dextran, casein, pullulan, gelatin, pectin, agar, carrageenan, xanthan gum, tragacanth, guar gum, acacia gum, arabic gum, polyethylene glycol, polyethylene oxide, polyvinyl pyrrolidone, polyvinyl alcohol, cyclodextrin, carboxyvinyl polymers, sodium alginate, polyacrylic acid, methylmethacrylate or mixtures thereof. In exemplary embodiments, the concentration of the water-soluble polymer in the formulation may be about 20% to about 90% (by weight), preferably between about 40% to about 80% (by weight).

[000131] In some embodiments, the pH sensitive polymer is Eudagrit® L100 that has a threshold pH (also called dissolution pH) of 6.0. In some embodiments, the pH sensitive polymer is Eudagrit® S100 that has a threshold pH of 7.0. In some embodiments, the pH sensitive polymer is Eudagrit® L-30D that has a threshold pH of 5.6. In some embodiments, the pH sensitive polymer is Eudagrit® FS 30D that has a threshold pH of 6.8. In some embodiments, the pH sensitive polymer is Eudagrit® L100-55 that has a threshold pH of 5.5. In some embodiments, the pH sensitive polymer is Polyvinyl acetate phthalate that has a threshold pH of 5.0. In some embodiments, the pH sensitive polymer is Hydroxypropylmethylcellulose phthalate that has a threshold pH of 4.5-4.8. In some embodiments, the pH sensitive polymer is Hydroxypropylmethylcellulose phthalate 50 that has a threshold pH of 5.2. In some embodiments, the pH sensitive polymer is

Hydroxypropylmethylcellulose phthalate 55 that has a threshold pH of 5.4. In some embodiments, the pH sensitive polymer is Cellulose acetate trimellitate that has a threshold pH of 4.8. In some embodiments, the pH sensitive polymer is Cellulose acetate phthalate that has a threshold pH of 5.0. In some embodiments the delayed release composition comprises a combination of the pH sensitive polymers mentioned above.

[000132] One skilled in the art, with the benefit of this disclosure, will understand that other components may be included to enhance one or more properties of the delayed release composition. In some embodiments, for example, the delayed release compositions may include one or more disintegrants, lubricants, anti-caking additives, anti-microbial agents, antifoaming agents, emulsifiers, surfactants, buffering agents, and/or coloring agents.

[000133] Suitable disintegrants include, for example, agar-agar, calcium carbonate, microcrystalline cellulose, croscarmellose sodium, crospovidone, povidone, polacrillin potassium, sodium starch glycolate, potato or tapioca starch, other starches, pre-gelatinized starch, clays, other alginates, other celluloses, gums, and mixtures thereof. In some embodiments, the disintegrant is crospovidone. In some embodiments, the disintegrant is croscarmellose sodium.

[000134] Suitable lubricants include, for example, calcium stearate, magnesium stearate, mineral oil, light mineral oil, glycerin, sorbitol, mannitol, polyethylene glycol, other glycols, stearic acid, sodium lauryl sulfate, talc, hydrogenated vegetable oil (e.g., peanut oil, cottonseed oil, sunflower oil, sesame oil, olive oil, corn oil and soybean oil), zinc stearate, ethyl oleate, ethyl laurate, agar, syloid silica gel (AEROSIL® 200, W.R. Grace Co., Baltimore, MD USA), a coagulated aerosol of synthetic silica (Evonik Degussa Co., Plano, TX USA), a pyrogenic silicon dioxide (CAB-O-SIL, Cabot Co., Boston, MA USA), and mixtures thereof.

[000135] Suitable anti-caking additives include, for example, calcium silicate, magnesium silicate, silicon dioxide, colloidal silicon dioxide, talc, glyceryl, and mixtures thereof.

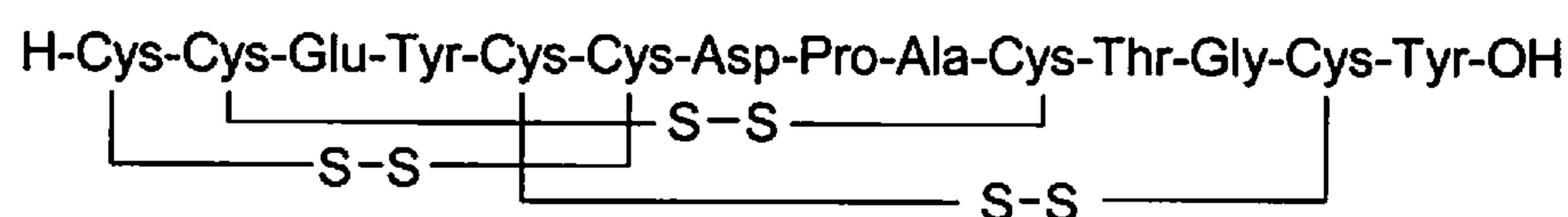
[000136] Suitable anti-microbial additives that may be used, *e.g.*, as a preservative for the linacotide compositions, include, for example, benzalkonium chloride, benzethonium chloride, benzoic acid, benzyl alcohol, butyl paraben, cetylpyridinium chloride, cresol, chlorobutanol, dehydroacetic acid, ethylparaben, methylparaben, phenol, phenylethyl alcohol, phenoxyethanol, phenylmercuric acetate, phenylmercuric nitrate, potassium sorbate, propylparaben, sodium benzoate, sodium dehydroacetate, sodium propionate, sorbic acid, thimersol, thymol, and mixtures thereof.

[000137] The composition may also comprise any suitable pharmaceutically acceptable carrier or medium. Suitable pharmaceutically acceptable carriers include, for example, any

solvents, dispersants, pH buffering agents, coatings, absorption promoting agents, controlled release agents, and one or more inert excipients (*e.g.*, filling agents, starches, polyols, granulating agents, microcrystalline cellulose, diluents, lubricants, binders, disintegrating agents), or the like. In addition, the compositions can contain any desired additional components, additives, and/or species, for example, surface active additives, dispersing additives, humectants, suspending agents, solubilizers, buffering agents, disintegrants, preservatives, colorants, flavorants, and the like. In some embodiments, the composition comprises one or more ion species that interact with linaclotide.

[000138] The composition can also comprise any suitable pH buffering agent. In some embodiments, the pH buffering agent is present in the composition in an amount sufficient to achieve the isoelectric point of linaclotide. In the regard, the composition can have any desired pH. In some embodiments, the composition has a pH of 2 to 5 (for example, a pH of 2 to 4.5, a pH of 2 to 4, a pH of 2.5 to 4, a pH of 2.5 to 3.5, a pH of 2.5 to 3, or even a pH of 3).

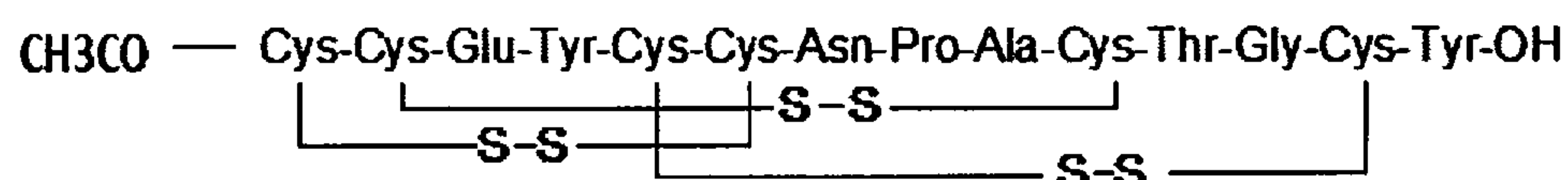
[000139] In some embodiments, the composition comprises linaclotide and a hydrolysis product, *e.g.*, a hydrolysis product comprising or having a structure of:



The composition can contain any desired concentration of the hydrolysis product. In some embodiments, the composition comprises less than 10 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 7 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 6 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 5 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 4 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 3 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 2 wt.% of the hydrolysis product. In some embodiments, the composition comprises less than 1 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 0.01 and 10 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 0.1 and 7 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 0.1 and 5 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 0.5 and 5 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 1 and 5 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 0.1 and 4 wt.% of the hydrolysis product. In some embodiments, the composition comprises between 0.5 and 4 wt.% of the hydrolysis product. In some embodiments, the

1 and 7 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 5 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 5 wt.% of the oxidation product. In some embodiments, the composition comprises between 1 and 5 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 4 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 4 wt.% of the oxidation product. In some embodiments, the composition comprises between 1 and 4 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 3 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 3 wt.% of the oxidation product. In some embodiments, the composition comprises between 1 and 3 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 2.5 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 2.5 wt.% of the oxidation product. In some embodiments, the composition comprises between 1 and 2.5 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 2 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 2 wt.% of the oxidation product. In some embodiments, the composition comprises between 1 and 2 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 1.5 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 1.5 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.1 and 1 wt.% of the oxidation product. In some embodiments, the composition comprises between 0.5 and 1 wt.% of the oxidation product.

[000141] In some embodiments, the composition comprises linaclootide and an acetylation product, *e.g.*, an acetylation product comprising or having:

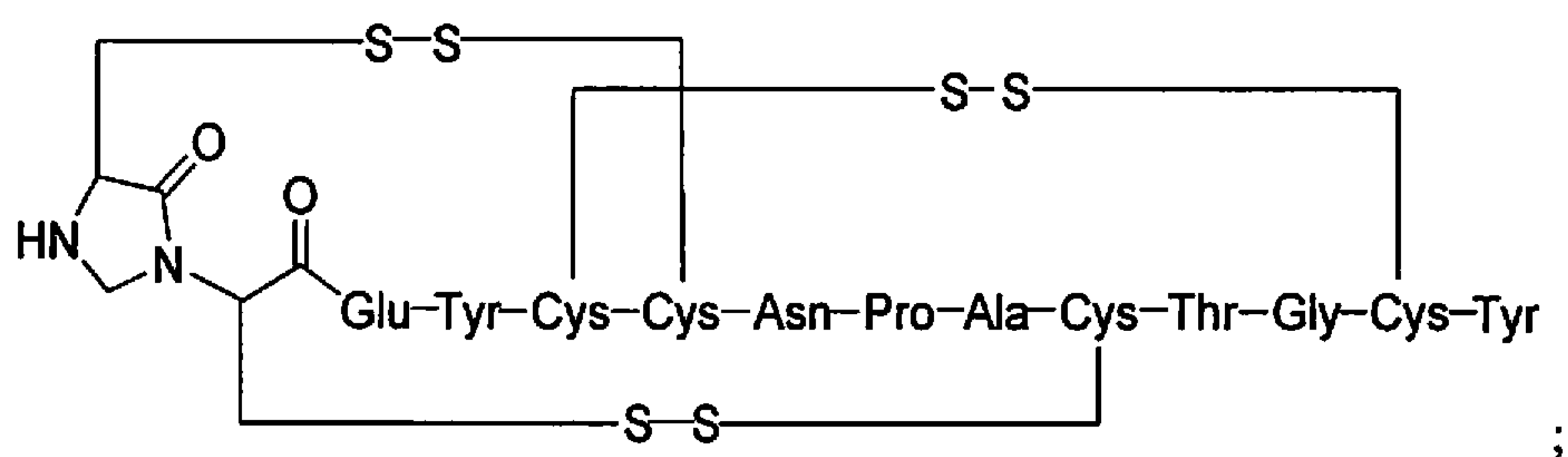


The composition can contain any desired concentration of the acetylation product. In some embodiments, the composition comprises less than 10 wt.% of the acetylation product. In some embodiments, the composition comprises less than 7 wt.% of the acetylation product. In some embodiments, the composition comprises less than 6 wt.% of the acetylation product. In some embodiments, the composition comprises less than 5 wt.% of the acetylation product. In some embodiments, the composition comprises less than 4 wt.% of the acetylation product. In some embodiments, the composition comprises less than 3 wt.% of the acetylation product.

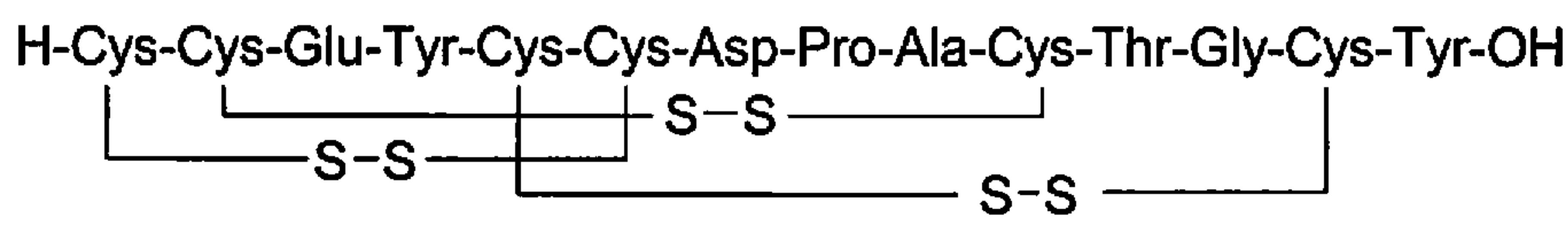
In some embodiments, the composition comprises less than 2 wt.% of the acetylation product. In some embodiments, the composition comprises less than 1 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.01 and 10 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 7 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 5 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 5 wt.% of the acetylation product. In some embodiments, the composition comprises between 1 and 5 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 4 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 4 wt.% of the acetylation product. In some embodiments, the composition comprises between 1 and 4 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 3 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 3 wt.% of the acetylation product. In some embodiments, the composition comprises between 1 and 3 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 2.5 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 2.5 wt.% of the acetylation product. In some embodiments, the composition comprises between 1 and 2.5 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 2 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 2 wt.% of the acetylation product. In some embodiments, the composition comprises between 1 and 2 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 1.5 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 1.5 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.1 and 1 wt.% of the acetylation product. In some embodiments, the composition comprises between 0.5 and 1 wt.% of the acetylation product.

[000142] In some embodiments, there is provided a pharmaceutical composition comprising linacotide, and one or more peptides selected from:

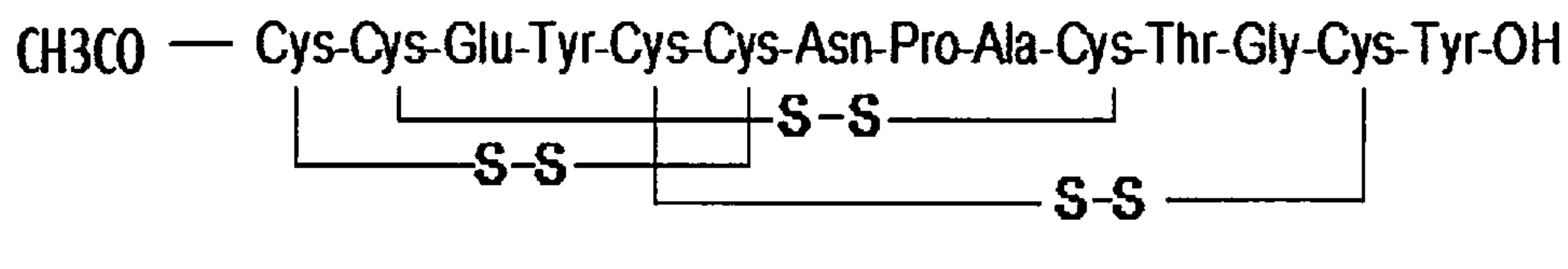
- i. a peptide (“Cys¹-IMD”) or a pharmaceutically acceptable salt thereof, wherein the peptide comprises the amino acid structure of:



- ii. a hydrolysis peptide ("Asp⁷") or a pharmaceutically acceptable salt thereof, wherein the peptide comprises the amino acid structure of:

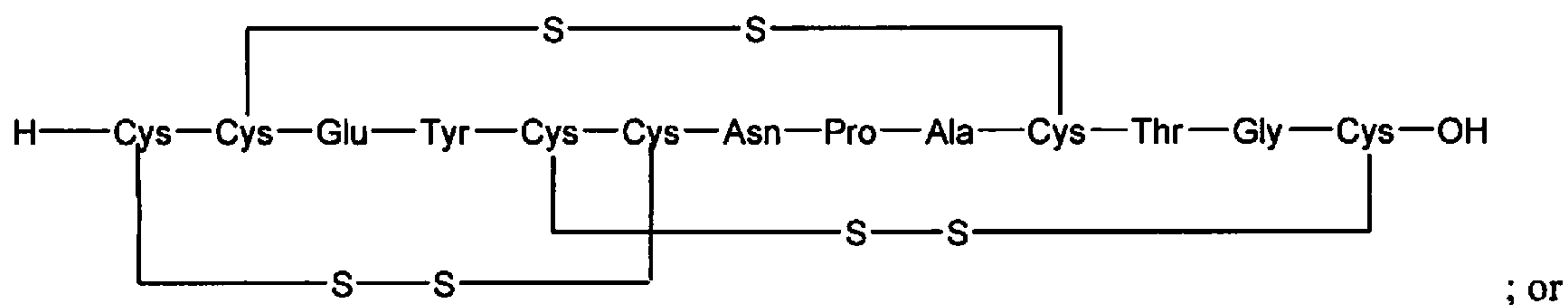


- iii. an acetylation peptide ("Cys¹-N-Acetyl") or a pharmaceutically acceptable salt thereof, wherein the peptide comprises the amino acid structure of:

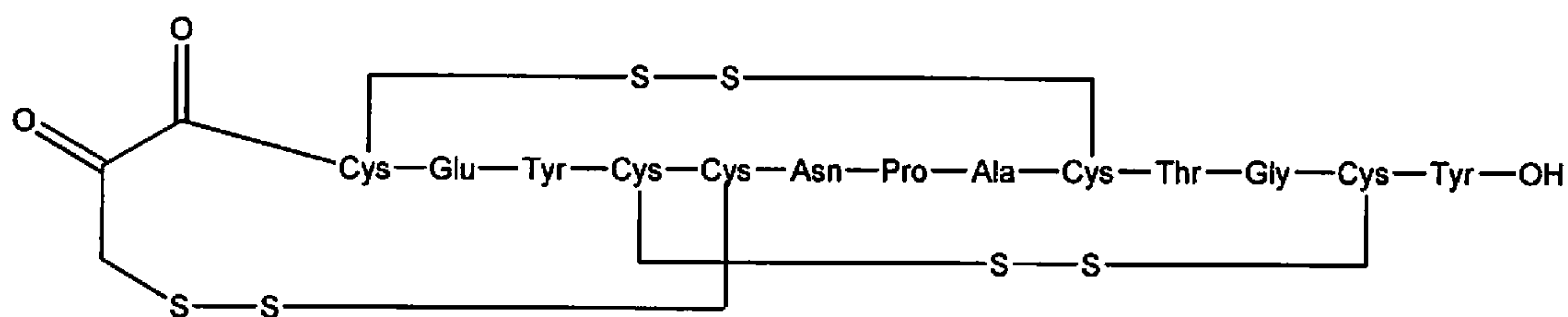


- iv. a linacotide trisulfide peptide or a pharmaceutically acceptable salt thereof, wherein the peptide comprises the amino acid sequence of Cys Cys Glu Tyr Cys Cys Asn Pro Ala Cys Thr Gly Cys Tyr wherein an additional sulfur atom may be attached to any one of the six cysteinyl sulfurs;

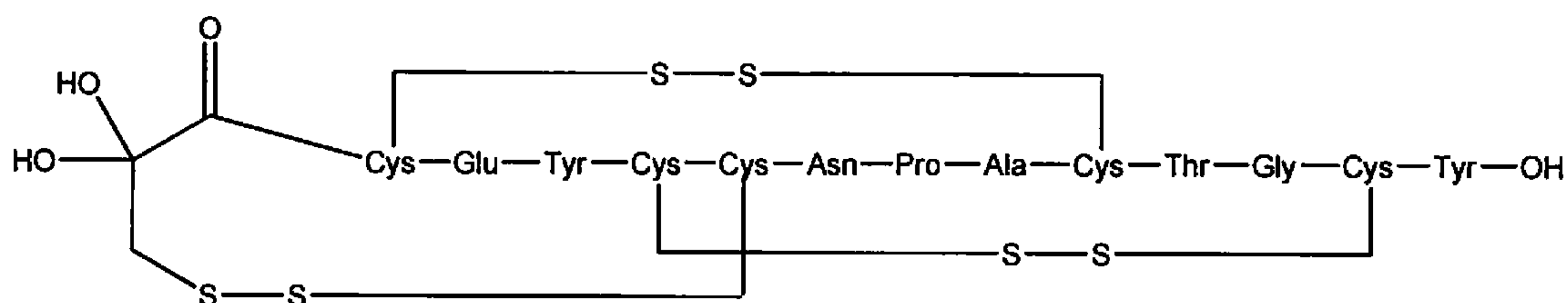
- v. a peptide ("Des-Tyr¹⁴") or a pharmaceutically acceptable salt thereof, wherein the peptide comprises the amino acid structure of:



- vi. a peptide (Cys¹-α-Ketone) or a pharmaceutically acceptable salt thereof, wherein the peptide comprises the amino acid structure of:



[000143] In some embodiments, the Cys¹-α-Ketone peptide may be present in its hydrated form or a pharmaceutically acceptable salt thereof, wherein the peptide comprises an amino acid structure of:



. One skilled in the art would recognize that the Cys¹-α-Ketone peptide would readily convert between its hydrate and ketone form.

[000144] In some embodiments, the Cys¹-α-Ketone peptide comprises less than about 15% by weight of the composition, less than about 10% by weight of the composition, less than about 7% by weight of the composition, less than about 5% by weight of the composition, less than about 4% by weight of the composition, less than about 3% by weight of the composition, less than about 2% by weight of the composition, less than about 1.5% by weight of the composition, or less than about 1% by weight of the composition. In other exemplary embodiments, the Cys¹-α-Ketone peptide comprises from about 0.01% to about 15% by weight of the composition, about 0.05% to about 10% by weight of the composition, about 0.05% to about 7% by weight of the composition or about 0.05% to about 5% by weight of the composition.

[000145] In some embodiments, the Cys¹-IMD peptide comprises less than about 15% by weight of the composition, less than about 10% by weight of the composition, less than about 7% by weight of the composition, less than about 5% by weight of the composition, less than about 4% by weight of the composition, less than about 3.5% by weight of the composition, less than about 3% by weight of the composition, less than about 2% by weight of the composition, or less than about 1% by weight of the composition. In other exemplary embodiments, the Cys¹-IMD peptide comprises from about 0.01% to about 15% by weight of the composition, about 0.05% to about 10% by weight of the composition, about 0.05% to about 7% by weight of the composition or about 0.05% to about 5% by weight of the composition.

[000146] In some embodiments, the hydrolysis peptide (“Asp⁷”) comprises less than about 15% by weight of the composition, less than about 10% by weight of the composition, less than about 7% by weight of the composition, less than about 5% by weight of the composition, less than about 4% by weight of the composition, less than about 3.5% by weight of the composition, less than about 3% by weight of the composition, less than about 2% by weight of the composition, or less than about 1% by weight of the composition. In other exemplary embodiments, the hydrolysis peptide (“Asp⁷”) comprises from about 0.01% to about 15% by weight of the composition, about 0.05% to about 10% by weight of the composition, about 0.05% to about 7% by weight of the composition or about 0.05% to about 5% by weight of the composition.

[000147] In some embodiments, the acetylation peptide (“Cys¹-N-Acetyl”) comprises less than about 15% by weight of the composition, less than about 10% by weight of the composition, less than about 7% by weight of the composition, less than about 5% by weight of the composition, less than about 4% by weight of the composition, less than about 3.5% by weight of the composition, less than about 3% by weight of the composition, less than about 2% by weight of the composition, or less than about 1% by weight of the composition. In other exemplary embodiments, the acetylation peptide (“Cys¹-N-Acetyl”) comprises from about 0.01% to about 15% by weight of the composition, about 0.05% to about 10% by weight of the composition, about 0.05% to about 7% by weight of the composition or about 0.05% to about 5% by weight of the composition.

[000148] In some embodiments, the linacotide trisulfide peptide comprises less than about 15% by weight of the composition, less than about 10% by weight of the composition, less than about 7% by weight of the composition, less than about 5% by weight of the composition, less than about 4% by weight of the composition, less than about 3.5% by weight of the composition, less than about 3% by weight of the composition, less than about 2% by weight of the composition, or less than about 1% by weight of the composition. In other exemplary embodiments, the linacotide trisulfide peptide comprises from about 0.01% to about 15% by weight of the composition, about 0.05% to about 10% by weight of the composition, about 0.05% to about 7% by weight of the composition or about 0.05% to about 5% by weight of the composition.

[000149] In some embodiments, the Des-Tyr¹⁴ peptide comprises less than about 15% by weight of the composition, less than about 10% by weight of the composition, less than about 7% by weight of the composition, less than about 5% by weight of the composition, less than about 4% by weight of the composition, less than about 3.5% by weight of the composition, less than about 3% by weight of the composition, less than about 2% by weight of the

composition, or less than about 1% by weight of the composition. In other exemplary embodiments, the Des-Tyr¹⁴ peptide comprises from about 0.01% to about 15% by weight of the composition, about 0.05% to about 10% by weight of the composition, about 0.05% to about 7% by weight of the composition or about 0.05% to about 5% by weight of the composition.

[000150] In some embodiments, the composition comprises linaclotide and any desired concentration of multimers. In some embodiments, the composition comprises less than 10 wt.% of multimer(s). In some embodiments, the composition comprises between 0.5 and 1 wt.% of multimer(s).

[000151] In some embodiments, the composition comprises an effective amount of linaclotide and any desired amount of reduced form linaclotide. As used herein, the term “reduced form linaclotide” refers to linaclotide having no disulfide bonds between cysteine amino acids. In some embodiments, the composition comprises less than 10 wt.% of reduced form linaclotide. In some embodiments, the composition comprises between 0.5 and 1 wt.% of reduced form linaclotide.

[000152] In some embodiments, the composition comprises an effective amount of linaclotide and any desired amount of scrambled form linaclotide. As used herein, the term “scrambled form linaclotide” refers to linaclotide having disulfide bonds between Cys₁ and Cys₁₀, between Cys₁ and Cys₁₃, between Cys₁ and Cys₅, between Cys₁ and Cys₂, between Cys₂ and Cys₆, between Cys₂ and Cys₁₃, between Cys₂ and Cys₅, between Cys₅ and Cys₆, and/or between Cys₅ and Cys₁₀. In some embodiments, the composition comprises between 0.5 and 1 wt.% of scrambled form linaclotide. In some embodiments, the composition comprises less than 10 wt.% of scrambled form linaclotide.

[000153] In some embodiments, the composition comprises a total degradant concentration of less than about 10 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 8 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 7 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 6.5 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 6 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 5.5 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 5 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 4 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 3 wt.%. In some embodiments, the composition comprises a total degradant concentration of

less than about 2.5 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 2 wt.%. In some embodiments, the composition comprises a total degradant concentration of less than about 1 wt.%.

[000154] In some embodiments, the compositions can be prepared by spray drying, which is a technique used to prepare microparticles (e.g., microcapsules or microspheres) of drugs. Spray-dried peptides generally retain their biological activity upon dissolution and may have useful physical characteristics, including a uniform particle size and a spherical shape. In addition, the microparticles prepared by spray drying are often free flowing, which is helpful for pharmaceutical manufacturing processes such as forming tablets and filling capsules. Spray drying processes are also useful because they may be readily scaled up for clinical and commercial manufacturing. In one embodiment, the spray buffer comprises HCl, histidine, 1.5% PVA and 0.6% talc. This formulation can be used to produce lower dosing ranges between 36-290µg.

[000155] The composition, when administered, will dissolve to release linaclotide in targeted areas of the gastrointestinal tract. The formulation may release the linaclotide over a period of time that is determined by a number of different factors. These factors include the dimensions of the formulation, the concentration of the linaclotide, and how the linaclotide is dispersed throughout the formulation. For example, by varying the thickness and surface area of the formulations the rate of dissolution may be adjusted. A thick formulation will dissolve more slowly than an otherwise similar thin formulation and may be desirable to administer high dosages of linaclotide.

[000156] In some embodiments, the delayed release composition has a disintegration rate of less than about 60 minutes in the targeted pH conditions. In some embodiments, the delayed release composition has a disintegration rate of less than about 30 minutes in the targeted pH conditions. In some embodiments, the delayed release composition has a disintegration rate of less than about 25 minutes. In some embodiments, the delayed release composition has a disintegration rate of less than about 20 minutes. In some embodiments, the delayed release composition has a disintegration rate of less than about 15 minutes. In some embodiments, the delayed release composition has a disintegration rate of less than about 10 minutes. In some embodiments, the delayed release composition disintegrates in less than about 30 minutes after entering a targeted environment. In some embodiments, the delayed release composition disintegrates in less than about 25 minutes after entering a targeted environment. In some embodiments, the delayed release composition disintegrates in less than about 20 minutes after entering a targeted environment. In some embodiments, the delayed release composition disintegrates in less than about 15 minutes after entering a targeted environment.

[000157] In some embodiments, the delayed release composition releases at least about 75% of the linaclotide contained therein within 60 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 75% of the linaclotide contained therein within 30 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 80% of the linaclotide contained therein within 30 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 85% of the linaclotide contained therein within 30 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 90% of the linaclotide contained therein within 30 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 95% of the linaclotide contained therein within 30 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 99% of the linaclotide contained therein within 30 minutes of entering a targeted environment.

[000158] In some embodiments, the delayed release composition releases at least about 40% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 50% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 60% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 70% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 80% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 85% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 90% of the linaclotide contained therein within 15 minutes of entering a targeted environment. In some embodiments, the delayed release composition releases at least about 95% of the linaclotide contained therein within 15 minutes of entering a targeted environment.

[000159] In some embodiments, the delayed release composition releases at least about 80% of the linaclotide contained therein between about 2 to about 2 hours of entering a targeted environment.

[000160] In some embodiments, the delayed release composition releases at least about 75% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 5.

In some embodiments, the delayed release composition releases at least about 80% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 5. In some embodiments, the delayed release composition releases at least about 85% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 5. In some embodiments, the delayed release composition releases at least about 90% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 5. In some embodiments, the delayed release composition releases at least about 95% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 5. In some embodiments, the delayed release composition releases at least about 99% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 5.

[000161] In some embodiments, the delayed release composition releases at least about 75% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 7. In some embodiments, the delayed release composition releases at least about 80% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 7. In some embodiments, the delayed release composition releases at least about 85% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 7. In some embodiments, the delayed release composition releases at least about 90% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 7. In some embodiments, the delayed release composition releases at least about 95% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 7. In some embodiments, the delayed release composition releases at least about 99% of the linaclotide contained therein within 30 minutes of contacting a pH greater than 7.

[000162] In some embodiments, the linaclotide DR compositions are formulated for delivery of linaclotide to the ileum, late ileum, or colon. In some embodiments, the linaclotide DR compositions are formulated for delivery of linaclotide to the ileum or ileal region. In some embodiments, the linaclotide DR compositions are formulated for delivery of linaclotide to within the late ileum to the ascending colon (e.g., at or near the ileocecal junction).

[000163] In some embodiments, the composition or oral dosage form is administered to a pediatric patient in need thereof as a tablet, capsule or sachet. In some embodiments, a sachet comprising the composition is opened and the contents are sprinkled on or stirred into food, such as applesauce, or into a beverage, such as water. In some embodiments, a capsule is swallowed whole with fluid, such as water, or is opened and sprinkled on or stirred into food or a beverage. Tablets may be swallowed whole, may be crushed and stirred into food or a beverage, or may be formulated as a chewable tablet.

[000164] A subject or patient in whom administration of the pharmaceutical composition is an effective therapeutic regimen for a disease or disorder is preferably a human, but can be any animal, including a laboratory animal in the context of a clinical trial or screening or activity experiment. Thus, as can be readily appreciated by one of ordinary skill in the art, the methods, compounds and compositions described herein are particularly suited for administration to any animal, particularly a mammal, and including, but by no means limited to, humans, rodents and non-rodents, such as feline or canine subjects, farm animals, such as but not limited to bovine, equine, caprine, ovine, and porcine subjects, wild animals (whether in the wild or in a zoological garden), research animals, such as mice, rats, rabbits, goats, sheep, pigs, dogs, cats, etc., avian species, such as chickens, turkeys, songbirds, etc., *e.g.*, for veterinary medical use.

[000165] In some embodiments, the linaclotide composition may be formulated as a rectal dosage form for rectal administration. Rectal dosage forms include, without limitation, rectal suppositories, rectal foams or aerosols, enemas, rectal gels and rectal ointments. In some embodiments, the rectal dosage form may be administered to a patient in need thereof. In some embodiments, the rectal dosage form may be administered to a pediatric or geriatric patient.

[000166] Another aspect of the invention is a method of making a delayed release composition, the method comprising: preparing a linaclotide base, pregranulated filler, and placebo base; and blending and compressing the linaclotide base, pregranulated filler, and placebo base into a tablet. In some embodiments, the pregranulated filler is prepared through wet granulation and dried before blending and compressing into a tablet. In some embodiments, the method further comprises applying a subcoat to the tablet. In some embodiments, the method further comprises applying an enteric or functional coating to the tablet.

[000167] Another aspect of the invention is a method of making a delayed release composition, the method comprising: preparing an aqueous solution comprising linaclotide, or a pharmaceutically acceptable salt thereof; and applying the aqueous solution to a pharmaceutically acceptable carrier.

Definitions

[000168] As used herein, unless otherwise indicated, the term “delayed release” mean that the composition dissolves, melts, disintegrates, liquefies, etc. in a targeted area of the gastrointestinal tract such that substantially all of the linaclotide no longer remains in a formulation, composition, or dosage form. Delayed release compositions include sustained release compositions, gastro-retentive compositions, targeted release compositions (*e.g.*

colonic-release compositions, or compositions that target the ileocecal valve, etc.), extended release compositions and/or combinations thereof.

[000169] As used herein, unless otherwise indicated, the term “delayed release composition” (“DR”) means that the composition is a dosage form that releases linaclotide at a time other than immediately following oral administration.

[000170] As used herein, unless otherwise indicated, the term “extended release composition” means that the composition is a dosage form that releases linaclotide over an extended period of time after administration. This allows a reduction in dosing frequency compared to immediate release compositions.

[000171] As used herein, unless otherwise indicated, the “disintegration” and “release” is used herein to mean that the capsule, film, bead, or tablet comprising linaclotide dissolves, melts, disintegrates, liquefies, etc. in the environment of an oral cavity such that substantially all of the linaclotide no longer remains in a formulation form, e.g., a pH greater than 5 or 7, or in a phosphate buffer solution and maintained at $37 \pm 1^\circ\text{C}$.

[000172] The term “released from”, when referring to the release of linaclotide from the composition, unless otherwise indicated, is used herein to mean that the linaclotide no longer remains in a composition form.

[000173] As used herein, unless otherwise indicated, the term “entry into a targeted environment” means contact of the composition within a patient at a targeted organ or segment thereof, or within a segment of the GI intended for linaclotide release, e.g., having a pH greater than 5 or 7.

[000174] As used herein, unless otherwise indicated, the term “lower gastrointestinal (GI)” means the distal segment of the gastrointestinal tract, for example, the ileum, terminal ileum, ileocecal valve, or colon.

[000175] As used herein, unless otherwise indicated, the term “upper gastrointestinal (GI)” means the proximate segment of the gastrointestinal tract, for example, the stomach, duodenum and/or jejunum.

[000176] As used herein, unless otherwise indicated, “stabilizing agent” refers to a polymer, sterically hindered primary amine (e.g., amino acid), or cation (e.g., metal cation) component of the composition which is included in the composition in a stabilizing amount. For example, a polymeric stabilizing agent is a polymer that is included in the composition in a stabilizing amount. Similarly, a sterically hindered primary amine stabilizing agent is a sterically hindered primary amine that is included in the composition in a stabilizing amount.

Moreover, a cationic stabilizing agent is a cation that is included in the composition in a stabilizing amount.

[000177] As used herein, unless otherwise indicated, “stabilizing amount” refers to a concentration, within the composition, of a polymer, sterically hindered primary amine (*e.g.*, amino acid), or metal cation component at which the component increases the stability of linaclotide in the composition, as compared to a similar composition not having a stabilizing amount of the same component.

[000178] As used herein, unless otherwise indicated, the term “substantially all” means at least about 90%, for example, at least about 95% or even at least about 99%.

[000179] As used herein, unless otherwise indicated, the term “isolated and purified” means at least 95 percent pure (for example, at least 96% pure, at least 97% pure, at least 98% pure, or even at least 99% pure), as measured, for example, by chromatographic purity using HPLC.

[000180] As used herein, unless otherwise indicated, “therapeutically effective amount” means the amount of a linaclotide or a pharmaceutically acceptable salt thereof that, when administered to a mammal for treating a state, disorder or condition, is sufficient to effect a treatment (as defined below). The “therapeutically effective amount” will vary depending on the compound, the disease and its severity and the age, sex, weight, physical condition and responsiveness of the mammal to be treated. For example, a therapeutically effective amount of linaclotide, or its pharmaceutically acceptable salt or hydrate, can be an amount effective to treat gastrointestinal disorders, including irritable bowel syndrome with constipation.

[000181] As used herein, unless other indicated, “pharmaceutically acceptable” means biologically or pharmacologically compatible for *in vivo* use in animals or humans, and preferably means, approved by a regulatory agency of the Federal or a state government or listed in the U.S. Pharmacopeia or other generally recognized pharmacopeia for use in animals, and more particularly in humans.

[000182] As used herein, unless otherwise indicated, the term “treat”, in all its verb forms, is used herein to mean to relieve, alleviate, prevent, and/or manage at least one symptom of a disorder in a subject, the disorder including, for example, a gastrointestinal disorder, such as irritable bowel syndrome with constipation. Within the meaning of the present invention, the term “treat” also denotes, to arrest, delay the onset (*i.e.*, the period prior to clinical manifestation of a disease) and/or reduce the risk of developing or worsening a disease. The term “treatment” means the act of “treating” as defined above.

[000183] As used herein, unless otherwise indicated, the term “prevent” refers to the prophylactic treatment of a subject who is at risk of developing a condition (e.g., stress related disorder) resulting in a decrease in the probability that the subject will develop the condition.

[000184] As used herein, unless otherwise indicated, the term “adverse event” refers to any untoward medical occurrence in a patient or clinical investigation subject administered a pharmaceutical product and which does not necessarily have a causal relationship with the treatment. For example, one adverse event is diarrhea.

[000185] As used herein, unless otherwise indicated, the term “additives” refers to a pharmaceutically acceptable additive. Pharmaceutically acceptable additives include, without limitation, binders, disintegrants, dispersing additives, lubricants, glidants, antioxidants, coating additives, diluents, surfactants, flavoring additives, humectants, absorption promoting additives, controlled release additives, anti-caking additives, anti-microbial agents (e.g., preservatives), colorants, desiccants, plasticizers and dyes.

[000186] As used herein, unless otherwise indicated, an “excipient” is any pharmaceutically acceptable additive, filler, binder or agent.

[000187] As used herein, unless otherwise indication, “stressed conditions” refer to 40°C and 75% relative humidity (RH).

[000188] As used here, unless otherwise indicated, the terms “about” and “approximately” mean within an acceptable error range for the particular value as determined by one of ordinary skill in the art, which will depend, in part, on how the value is measured or determined, *i.e.*, the limitations of the measurement system. For example, “about” can mean within 1 or more than 1 standard deviation, per practice in the art. Alternatively, “about” with respect to the compositions can mean plus or minus a range of up to 20%, preferably up to 10%. Alternatively, particularly with respect to biological systems or processes, the term can mean within an order of magnitude, preferably within 5-fold, and more preferably within 2-fold, of a value. Particular values are described in the application and claims, unless otherwise stated the term “about” means within an acceptable error range for the particular value.

[000189] All weight percentages (*i.e.*, “% by weight” and “wt.%” and w/w) referenced herein, unless otherwise indicated, are measured relative to the total weight of the pharmaceutical composition.

[000190] The term “consisting essentially of”, and variants thereof, when used to refer to the composition, are used herein to mean that the composition includes linaclotide and other desired pharmaceutically inactive additives, excipients, and/or components (*e.g.*, polymers, sterically hindered primary amines, cations, filling agents, binders, carriers, excipients, diluents, disintegrating additives, lubricants, solvents, dispersants, coating additives,

absorption promoting additives, hydrolysis products, formaldehyde imine products, oxidation products, acetylation products, deamidation products, multimers, controlled release additives, anti-caking additives, anti-microbial additives, preservatives, sweetening additives, colorants, flavors, desiccants, plasticizers, dyes, or the like), and no other active pharmaceutical ingredient(s).

EXAMPLES

[000191] The following examples are merely illustrative of the present invention and should not be construed as limiting the scope of the invention in any way as many variations and equivalents that are encompassed by the present invention will become apparent to those skilled in the art upon reading the present disclosure.

[000192] Enteric-coated tablets comprising a core immediate release tablet containing a unit dose of linaclotide can be coated with coatings that dissolve only under pH conditions of the distal segment of intestine, so that linaclotide will be released in lower GI tract.

[000193] Linaclotide or a pharmaceutically acceptable salt thereof may be produced and purified using standard techniques known in the art, e.g., chemical synthesis or recombinant expression followed by purification using standard techniques.

[000194] Preparation of the linaclotide coating solution for beads: Approximately 32 g to 42 g of purified water is mixed with hydrochloric acid to create a solution with a pH between 1.5 and 2.0. The cation, if used, is added to the solution in a quantity to provide the desired concentration, and the solution is mixed for sufficient time to produce a clear solution. The sterically hindered primary amine, if used, is added to the solution in a quantity to provide the desired concentration, and the solution is mixed for sufficient time to produce a clear solution. Other additives, such as antioxidants, are then added, if desired. The pH of the solution is tested, and hydrochloric acid is added, if necessary, to produce a solution having a pH between 1.5 and 2.0. The binder is then added to the solution and the mixture is then stirred for sufficient time to achieve a clear solution. The desired amount of linaclotide is added to the solution and mixed for 30-100 minutes to provide the coating solution.

[000195] In one embodiment, the coating solution comprises linaclotide, histidine, 1.5% PVA and 0.6% talc. This formulation can be used to produce dosing ranges between 30-300µg.

[000196] Preparation of the Active Beads: Approximately 30-36 g of dried microcrystalline cellulose beads are added to a Mini Column Fluid Bed Coater. The microcrystalline cellulose beads are fluidized and heated prior to layering. Next, the coating solution is layered to the beads. The spraying temperature is controlled between 24°C and 55°C by controlling inlet temperature, spray rate, atomization pressure, and air volume. After the

entire coating solution is layered to the beads, the beads are dried. The product of this process is referred to as active beads.

Example 1

Delayed release Linacotide Tablet

[000197] Linacotide can be formulated into a tablet for delayed drug release. Compared to an equal volume of beads, tablets have much smaller specific surface area, which makes them potentially less prone to degradation induced by environmental factors such as humidity, oxidation, deamidation, etc. In addition, the smaller surface area of the tablet can become advantageous when an enteric coating is needed since much less coating material is required to cover the surface of the dosage form.

[000198] Enteric coatings may be applied in a tablet coating pan, and coatings that are used for delayed release beads can be used for tablets to form delayed release tablets. The amount of coating polymer on the tablet can vary from 5 to 60% (weight gain) depending on the size, shape and surface properties of the tablet. A sub-coat can be applied to the tablets to separate linacotide from the enteric or functional coat.

Example 2

Enteric coated tablet

Table 1 Eudragit® FS30D Coated linacotide delayed release (DR) tablet composition

	Ingredients	Wt. %	Wt. in kg
Fluid bed Granulation			
1.	Linacotide	0.3	2.94
2	Isomalt	93.7	937
3	Histidine	0.46	4.6
4	Calcium chloride dihydrate	2.57	25.7
5	Polyvinyl pyrrolidone (PVP)	3	30
6	0.01N HCl	Q.S.	Q.S.
7	Purified Water	Q.S.	Q.S.
Blending and compression			

i	Linacotide granules	27.85	139.25
ii	Isomalt	60.9	304.5
iii	Crospovidone	10	50
iv	Magnesium stearate	0.75	3.75
vi	Talc	0.5	2.5
Enteric coating			
	Linacotide tablet	75.19	1000
	Eudragit® FS 30D	22.56	1000
	PlasACRYL™	2.25	150
	Purified Water*	-	500
	Total Dry weight	100	1330

Manufacturing process:

A. Tablet

[000199] The granulation solution may be prepared by dissolving PVP, histidine and calcium chloride in water, adjusting solution pH to 2, and dissolving linacotide. Granulation is performed in a fluid bed by spraying the granulation solution onto filler isomalt. At the end of granulation, dry the granules for 30 min. The granules are then blended with tablet components including isomalt, crospovidone, Mg stearate and talc until uniform, and compressed into tablets.

B. Enteric coating

[000200] For tablet coating, linacotide core tablets are placed into a pan coater and warmed up to 35°C. Start tablets coating with Eudragit® FS 30 D suspension, keep the product temperature at 28° to 32°C, and atomization air pressure at 3 bar. At the end of coating, discharge the tablets and place them into a circulated air oven and dry for 2 h at 40°C. Similarly, other enteric coatings such as Eudragit® L, S, ethyl cellulose, HPMCAS, PVAP, CAP, CAS, etc. may also be applied to form delayed release tablets at various weight gains.

Example 3

Delayed release compositions comprising linacotide

[000201] Delayed release capsules comprising linaclotide may be formulated to target the ileum or colon (e.g., the ileum, late ileum, and/or ascending colon). The composition is formulated to include a pH triggered release based on enteric coating of a linaclotide tablet, capsule or linaclotide coated beads contained in a hard gelatin capsule. The composition may be formulated to further comprise stabilizing additives such as a divalent cation and an amino acid. PVA can be used as binder as well as protective layer in between linaclotide and enteric coating. Linaclotide or linaclotide with PVA overcoat (as beads, capsule or tablet) may be coated with an additional enteric coating (e.g. Eudragit® FS30D, Eudragit® S100, Eudragit® L100, Eudragit® L100-55, Eudragit® L 30D-55) that dissolves in a pH dependent manner to release at the appropriate pH of 7 in the ileum of the GI tract. The enteric coatings may consist of blends combining different types of Eudragit® - Eudragit® S100 / Eudragit® L100 in different ratios (e.g. 50/50 ratio); Eudragit® S100 / Eudragit® L100-55 in various ratios; Eudragit® FS30D/Eudragit® L 30D-55, Eudragit® FS30D/Eudragit® S / Eudragit® RS or EC in various ratios. The compositions may further comprise other excipients including plasticizing agents such as triethylcitrate. The coatings may further comprising disintegrants as suspended solid to expedite the relevant pH triggered release – resulting in mixed systems as croscarmellose sodium / Eudragit® S. For ease of processing, anti-tacking agent (e.g., talc, Aerosil® 200 or PlasAcryl™) may be used to prevent the beads from sticking.

[000202] Additionally, two Eudragit® coatings may be applied to ensure swift release once the desired pH region in the GI tract is reached – including partially neutralized coating systems. Buffering agents such as potassium hydrogen phosphate can be included into one of the two Eudragit® films. Alternative non-Eudragit® pH dependent film coatings include hydroxypropylmethylcellulose acetate succinate (HPMCAS, e.g. Aqoat® AS-HF), cellulose acetate phthalate (CAP, e.g. Aquateric®) or shellac.

Example 4

Measurement of content and purity of exemplary peptides

[000203] Linaclotide, imidazolidinone degradant product (“Cys¹-IMD”), and α -ketone degradant product (“Cys¹- α -Ketone”) can be measured and purified as described in US 2010/0048489, US 2013/0190238, and US 2015/0094272 which are incorporated by reference herein. Generally, content and purity of linaclotide may be determined by reverse phase gradient liquid chromatography using an Agilent Series 1100 LC System with Chemstation Rev A.09.03 software or equivalent. A YMC Pro™ C18 column (dimensions: 3.0 x 150 mm,

3.5 μm , 120 $\text{\AA}$; Waters Corp., Milford, MA) or equivalent is used and is maintained at 40°C. Mobile phase A (MPA) consists of water with 0.1% trifluoroacetic acid while mobile phase B (MPB) consists of 95% acetonitrile:5% water with 0.1% trifluoroacetic acid. Elution of the linaclotide is accomplished with a gradient from 0% to 47% MPB in 28 minutes followed by a ramp to 100% MPB in 4 minutes with a 5 minute hold at 100% MPB to wash the column. Re-equilibration of the column is performed by returning to 0% MPB in 1 minute followed by a 10 minute hold at 100% MPA. The flow rate is 0.6 mL/min and detection is accomplished by UV at 220 nm.

Example 5

In vitro drug release testing

[000204] Linaclotide release from the coated tablets can be assessed by dissolution testing using a USP XXIV type II paddle dissolution apparatus (model PTWS, Pharma Test, Hainburg, Germany). The tests are conducted in triplicates, at a paddle speed of 100 rpm in 900 ml dissolution medium maintained at $37.0 \pm 0.5^\circ\text{C}$. Tablets are tested first in 0.1 N hydrochloric acid (pH 1.2) for 30 minutes or 2 hours to simulate gastric residence and then for 6 hours in buffer media of varying pH and ionic composition, akin to small intestinal pH conditions. Linaclotide release may be assessed at pH 6.8–7.4 in two compendial buffer media: 0.067 M mixed sodium and potassium phosphate (Sorensen's) buffer and 0.05 M potassium phosphate buffer as well as in a pH 7.4 multi-electrolyte salt solution (Hanks) buffer which is similar in ionic composition to intestinal fluid. All the buffers are freshly prepared with de-ionized water and de-aerated by sparging with helium prior to use. The amount of linaclotide released from the dosage form can be determined by reverse phase gradient liquid chromatography as described in Example 4.

Example 6

Linaclotide Tablet Release Profiles

[000205] Linaclotide tablet release profiles have been tested at various pHs for three delayed release formulations. Sample 181-64B contains a 100 μg dose of linaclotide, a subcoat of Opadry® II, and a functional coat of Eudragit® FS 30D and talc. Sample 181-64C contains a 100 μg dose of linaclotide, a subcoat of Opadry II, and a functional coat of Eudragit® FS 30D and Eudragit® PlasAcryl™. The release profiles for Lot 181-64B and Lot 181-64C are provided in Figure 20, top. In addition, various formulations were tested using varying ratios of Eudagrit L100 and Eudagrit S100, and tested using the Physiolution® dissolution device

(Physioloution GmbH, Greifswald, Germany). Briefly, various tablets were tested for drug dissolution in physiological buffers under conditions of varying pH mimicking gastrointestinal passage in order to determine the location of dissolution. As is shown in Figure 20B, the rate of drug release is plotted against time during which the device alters the pH according to Table 40.

Table 40.

time (h)	pH	Simulated Location
0.00	1.8	Stomach
0.17	1.8	Stomach
0.33	1.8	Stomach
0.48	1.8	Stomach
0.50	5.81	Duodenum/Jejunum
0.67	6.01	Duodenum/Jejunum
0.83	6.05	Duodenum/Jejunum
1.00	6.17	Duodenum/Jejunum
1.17	6.3	Duodenum/Jejunum
1.33	6.34	Duodenum/Jejunum
1.50	6.54	Duodenum/Jejunum
1.67	6.71	Duodenum/Jejunum
1.83	6.95	Ileum
2.00	7.1	Ileum
2.17	7.23	Ileum
2.33	7.36	Ileum
2.50	7.4	Ileum
2.67	7.48	Ileum
2.83	7.58	Ileum
3.00	7.57	Ileum
3.17	7.66	Ileum
3.33	7.66	Ileum/Colon junction
3.50	7.67	Ileum/Colon junction
3.67	7.69	Ileum/Colon junction
3.83	7.68	Ileum/Colon junction
4.00	7.65	Ileum/Colon junction
4.17	7.64	Ileum/Colon junction
4.33	7.43	Ileum/Colon junction
4.48	7.43	Ileum/Colon junction
4.50	6.96	Colon
4.67	7.85	Colon

4.83	6.88	Colon
5.00	6.96	Colon
5.17	6.79	Colon
5.33	6.76	Colon
5.50	6.76	Colon
5.67	6.55	Colon
5.83	6.34	Colon
6.00	6.35	Colon
6.17	6.69	Colon
6.33	6.34	Colon
6.50	6.34	Colon
6.67	6.96	Colon
6.83	7.17	Colon
7.00	6.88	Colon
7.17	6.96	Colon
7.33	6.55	Colon
7.50	6.76	Colon
7.67	6.44	Colon
7.83	6.55	Colon
8.00	6.96	Colon

In Figure 20B identical core tablets of linaclotide prepared as described herein were coated with different ratios of Eudagrit L100 (which has a dissolution above pH 6.0) and Eudragit S100 (with a dissolution above pH 7.0). By varying the ratios from 40:60 (S100:L100) to 95:5 (S100:L100), the release of the drug can be dramatically altered, from substantial release in conditions of the duodenum/jejunum (40:60, 50:50), the ileum (60:40, 80:20, and 85:15), to the lower ileum/colon (90:10, and 95:5). Based on these studies, ratios providing release in the upper (83:17 S100:L100) and lower (100% S100) parts of the lower GI were chosen for delayed release formulations.

Example 7

Linaclotide Tablet Preparation

[000206] Delayed release tablets may be prepared by first preparing the following core tablet components: a placebo base, a linaclotide 750 µg/225 mg base, and pre-granulated fillers.

Granulation manufacturing process:

[000207] The tablet components may be prepared into separate granulations for blending before tablet compression. Use of separate tablet components, such as, the placebo base and pregranulated filler base provided, among other things, advantageous properties for stability and release profiles for the tablets. For example, all the tablets components listed in Table 2 could be separately prepared by wet granulation and blended before compression or blended together and processed as a mixture for wet granulation. In another process, the tablets components listed in Table 2 could be separately prepared by dry granulation and blended before compression or blended together and processed as a mixture for dry granulation. In another process, the tablet components are direct blended for compression. In a preferred process, the pregranulated filler base and/or placebo base are prepared through wet granulation and dried before mixing with the 750 µg/225 mg linaclotide base. The linaclotide base could be prepared by wet granulation processes or by Wurster coating process. This preferred process, exhibited further gains in stability for the tablet by reducing moisture exposure to linaclotide during processing and minimizing residue moisture in the tablet core.

Table 2: Components for various tablet strengths

Strength	Placebo	25 µg	30µg	50 µg	75 µg	100 µg	150 µg	290 µg	300µg
Placebo base (%)	20.00	16.67	16	13.33	10.00	6.67	3.33	0.00	0
Linaclotide base (i.e.750 ug/225 mg base (%))	0.00	3.33	4	6.67	10.00	13.33	16.67	38.65	40
Pregranulated fillers (%)	78.75	78.75	78.75	78.75	78.75	78.75	78.75	60.10	58.75
Magnesium Stearate (%)	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25
Total (%)	100.0	100.0	100	100.0	100.0	100.0	100.0	100.0	100

[000208] Then compress the above blends on a suitable tablet press to target core tablet weight of 225 mg. In a perforated pan coater, add a sub-coat (OPADRY® II) at a weight gain of 4% w/w. Coating conditions should be set and monitored so that moisture uptake during coating is kept to a minimum. When measured by loss on drying (LOD), the sub-coated tablets

should have no more than 1.5% LOD. In a perforated pan coater, add a functional coat on the subcoated tablets. The functional coat is either Eudragit® FS30D, Eudragit® S100, or Eudragit® L100. Apply the functional coat at 5 mg polymer weight/cm² of the tablet surface. This comes to be approximately 4.5% total polymer weight gain during functional coating. Coating conditions should be set and monitored so that moisture uptake during coating is kept to a minimum. When measured by loss on drying, the functionally coated tablet should have no more than 2.0% LOD.

Placebo Base Preparation:

[000209] Table 3 represents the formulation for the placebo base granulation:

Table 3: Formulation of the placebo base granulation

Component	%w/w	Quantity (g)
L-Histidine	2.26	112.9
Calcium Chloride Dihydrate	1.07	53.5
polyvinyl alcohol	1.50	75.0
microcrystalline cellulose	95.17	4758.6
Hydrochloric acid, pure, fuming, 37% solution in water		
Treated water		
Total	100	5000

[000210] The placebo base preparation may be prepared by first dispensing the raw materials of Table 4.

Table 4: Raw materials for placebo base preparation

Component	Quantity (g)
L-Histidine	242.2
Calcium Chloride Dihydrate	114.8
polyvinyl alcohol	160.9
microcrystalline cellulose	4758.6

[000211] Tare the mix container and add 2682.1 ± 5.0 g of treated water into the container. Set up a mixer and begin to stir the water. Add the EMPROVE® to the water while stirring and start the timer. Cover and heat solution to 70C while stirring and maintain temperature until material is visually dissolved.

[000212] Adjust the pH of solution to 1.5 with hydrochloric acid. Add calcium chloride dihydrate to the solution while stirring. Mix until dissolved. Add L-Histidine to the solution while stirring. Stir for approximately 15 minutes. Record the initial pH. Adjust pH of solution to 5.0 with hydrochloric acid. Record final pH of solution and hydrochloric acid addition. Mix until all material is dissolved. While mixing, adjust the pH solution to 2.5 with hydrochloric acid. Record final pH of solution and hydrochloric acid addition. Ensure that the high shear granulator is set up properly for granulating with the 25L bowl, mixing blade and chopper. Pass microcrystalline cellulose through 16 mesh screen into granulator bowl. Calculate the net weight of granulation solution to add. Pump the granulation solution into the granulator at a rate of approximately 300g/min while mixing with the below parameters: Impeller speed 1(290 rpm, 5.5m/s tip speed), Chopper speed 1(1760 rpm). Stop the granulator and scrape down the sides and the bottom of the bowl. Mix for an additional 3 minutes according to the following parameters: Impeller speed 1(290 rpm, 5.5m/s tip speed), Chopper speed 1(1760 rpm). Tare a poly bag and discharge the completed wet granulation into it. Weigh the granulation. Transfer the wet granulation to the FLM-3 fluid bed for drying. Dry the granulation using the following approximate settings. Dry until the granulation LOD is no more than 1.2% moisture. Discharge the dried granulation into a tared poly bag.

Parameters	Target Range
Product Temperature	40°C
Process (Drying) Air	30 – 60 CFM
Inlet Temperature	60°C

Settings are suggested settings only and may be adjusted for optimum drying.

[000213] Screen the dried granulation through a #30 mesh sieve. Tare a poly bag and discharge the dried granulation into it. Weigh the granulation. Package dried granulation into foil sealed bags with desiccant.

Linacotide Base Preparation (i.e. 750µg/225mg):

[000214] Table 5 represents the formulation for the 750µg/225mg base granulation:

Table 5: Formulation for the 750µg/225mg base granulation

Component	% w/w	Quantity (g)
Linacotide	0.39	19.3
L-Histidine	2.26	112.9
Calcium Chloride Dihydrate	1.07	53.5
polyvinyl alcohol	1.50	75.0
microcrystalline cellulose	94.79	4,739.3

Hydrochloric acid, pure, fuming, 37% solution in water	-----	-----
Treated water	-----	-----
Total	100.00	5,000.0

[000215] The 750µg/225mg base granulation may be prepared by first dispensing the raw materials of Table 6.

Table 6: Raw materials of the linaclotide base granulation

Raw Material	Required Quantity (g)
Linaclotide	19.3
microcrystalline cellulose	4,739.3
Granulation solution	

[000216] While mixing, add the linaclotide to the granulation solution. Mix until dissolved. Ensure that the high shear granulator is set up properly for granulating with the 25L bowl, mixing blade and chopper. Pass microcrystalline cellulose through 16 mesh screen into granulator bowl. Pump the granulation solution into the granulator at a rate of approximately 300g/min while mixing with the below parameters: Impeller speed 1(290 rpm, 5.5m/s tip speed), Chopper speed 1(1760 rpm). Tare a poly bag and discharge the completed wet granulation into it. Weigh the granulation. Transfer the wet granulation to the fluid bed for drying. Dry the granulation using the following approximate settings. Dry until the granulation LOD is no more than 1.2% moisture. Discharge the dried granulation into a tared poly bag.

Parameters	Target Range
Process (Drying) Air	30 – 60 CFM
Inlet Temperature	80°C

Note: Settings are suggested settings only and may be adjusted for optimum drying.

[000217] Screen the dried granulation through a #30 mesh sieve. Tare a poly bag and discharge the dried granulation into it. Weigh the granulation. Package dried granulation into foil sealed bags with desiccant.

Pregranulated Filler Preparation:

[000218] Table 7 represents the formulation for the pregranulated fillers.

Table 7: Formulation of the fillers granulation

Component	% w/w	Qty per subplot (g)
microcrystalline cellulose	19.4	1,358

croscarmellose sodium	5.1	357
mannitol	71.7	5,019
polyvinyl alcohol	3.8	266
Treated water	-----	-----
Total	100.0	7,000

[000219] The fillers preparation may be prepared by first dispensing the raw materials of Table 8.

Table 8: Raw materials for preparation of fillers granulation

Raw Material	Required Quantity (g)
polyvinyl alcohol	266
microcrystalline cellulose	1,358
croscarmellose sodium	357
mannitol	5,019
Total	---

[000220] Then record the tare weight of the stainless steel container. Tare the container and weigh the required quantity of treated water into the container. Transfer the water into a jacketed kettle. Set up the mixer and begin to stir the water in the kettle. Add the EMPROVE® (polyvinyl alcohol) to the water while stirring and start the timer. Cover and heat solution to 70°C while stirring and maintain temperature until material is visually dissolved. Calculate weight of water lost due to evaporation during heating. Add this amount of treated water to the solution. Add each of microcrystalline cellulose, croscarmellose sodium), and mannitol to a high shear granulator bowl. Mix for approximately 2 minutes according to the following parameters: Impeller speed 1(290 rpm, 5.5m/s tip speed), Chopper speed 1(1760 rpm). Pump 2217 ± 5 g of the granulation solution into the granulator at a rate of approximately 300g/min while mixing with the below parameters: Impeller speed 1(290 rpm, 5.5m/s tip speed), Chopper speed 1(1760 rpm). Stop the granulator and scrape down the sides and the bottom of the bowl. Mix for an additional 30 seconds to 1 minute according to the following parameters: Impeller speed 1(290 rpm, 5.5m/s tip speed), Chopper speed 1(1760 rpm). Tare a poly bag and discharge the completed wet granulation into it. Weigh the granulation. Pass the wet granulation through the Comil with 2A375Q03763 screen with 5-10% power. Transfer the wet granulation to the FLM-3 fluid bed for drying. Dry the granulation using the following approximate settings. Dry until the granulation LOD is no more than 1.0% moisture. Discharge the dried granulation into a tared poly bag.

Parameters	Target Range
Process (Drying) Air	30 – 60 CFM
Inlet Temperature	80°C

Settings are suggested settings only and may be adjusted for optimum drying.

[000221] Mill the granulation with Comil, round impeller, 2A045R03137 screen. Tare a poly bag and discharge the dried and milled granulation into it. Weigh the granulation. Package dried granulation into foil sealed bags with desiccant.

Example 8

25µg Tablet Blending and Compression

[000222] According to the procedure of Example 7, a 25µg dose tablet of linacotide may be prepared with the formulation of Table 9.

Table 9: Formulation of a 25 µg Linacotide Tablet:

Component	% w/w	Quantity (g)
Pregranulated Fillers	78.750	5118.8
Linacotide Base	3.333	216.7
Placebo Base	16.667	1083.4
Magnesium Stearate	1.250	81.3
Total	100	6500

[000223] First, dispense the raw materials of Table 10.

Table 10: Raw materials for preparation of 25µg linacotide tablet

Raw Material	Required Quantity (g)
Pregranulated Fillers	5118.8
Linacotide Base	227.5
Placebo Base	1137.5
Magnesium Stearate	81.2
Total	6565.0

[000224] Preblend: Set up an 8 qt blender and add the 750µg/225mg Base and Placebo Base. Close lids and blend for 10 minutes. Tare a poly bag, and discharge the preblend into it.

[000225] Sub-Blend A: Add 650g of the preblend to a 16 qt v-blender. Add the Pregranulated Fillers for sub-blend A to the 16 qt v-blender. Close lids and blend for 10 minutes. Pass the Magnesium Stearate for sub-blend A through the 40 mesh screen. Add to the 16 qt v-blender. Close lids and blend for 3 minutes. Tare a poly bag, and discharge the blend into it.

[000226] Sub-Blend B: Add 650g of the preblend to a 16 qt v-blender. Add the Pregranulated Fillers for sub-blend B to the 16 qt v-blender. Close lids and blend for 10 minutes. Pass the Magnesium Stearate for sub-blend B through the 40 mesh screen. Add to the 16 qt v-blender. Close lids and blend for 3 minutes. Tare a poly bag, and discharge the blend into it.

[000227] Compression: Ensure a Korsch XL 100 tablet press with 0.32" sc toolings is set up with 0.32" round concave tooling (10 stations desired), paddle feeder, and that all other components are properly secured. Manually rotate die table at least one full revolution to ensure proper installation. Visually confirm presence of all dies and punches, and that dies are flush with the die table. Verify that the tablet specifications meet the In-Process Tablet Specifications of Table 11.

Table 11: In-Process Tablet Specifications

Parameters	Target
Average Tablet Weight	225 mg
Individual Thickness	4.0 - 4.5 mm (guideline only)
Individual Tablet Hardness	10 kP
Friability (100 drops)	NO MORE THAN 0.6%
Individual Disintegration	NO MORE THAN 15 min

[000228] Charge the blend into the hopper. Set the turret speed to an appropriate speed. Adjust the die fill amount and compression parameters to yield a tablet with a target weight of 225 mg and with a target hardness of 8-12 kP. All waste material should be collected in the tableting waste poly bag. Start timer and compress tablets.

Example 9

100µg Tablet Blending and Compression

[000229] According to the procedure of Example 7, a 100 μ g dose tablet of linaclotide may be prepared with the formulation of Table 12.

Table 12: Formulation of a 100 μ g Linaclotide Tablet:

Component	% w/w	Quantity (g)
Pregranulated Fillers	78.750	9843.8
Linaclotide Base	13.333	1666.6
Placebo Base	6.667	833.4
Magnesium Stearate	1.250	156.3
Total	100	12500

[000230] First, dispense the raw materials of Table 13.

Table 13: Raw materials for preparation of 100 μ g linaclotide tablet

Raw Material	Required Quantity (g)
Pregranulated Fillers	9843.8
Linaclotide Base	1750.0
Placebo Base	875.0
Magnesium Stearate	156.3
Total	12,625.0

[000231] Preblend: Set up an 16 qt blender and add the 750 μ g/225mg Base and Placebo Base. Close lids and blend for 10 minutes. Tare a poly bag, and discharge the preblend into it.

[000232] Add 2500g of the preblend to a 1 cubic foot v-blender. Add the Pregranulated Fillers to the 1 cubic foot v-blender. Close lids and blend for 10 minutes. Pass the Magnesium Stearate through the 40 mesh screen. Add to the 1 cubic foot v-blender. Close lids and blend for 3 minutes. Tare a poly bag, and discharge the blend into it.

[000233] Compression: Ensure a Korsch XL 100 tablet press with 0.32" sc toolings is set up with 0.32" round concave tooling (10 stations desired), paddle feeder, and that all other components are properly secured. Manually rotate die table at least one full revolution to ensure proper installation. Visually confirm presence of all dies and punches, and that dies are flush with the die table. Verify that the tablet specifications meet the In-Process Tablet Specifications of Table 14.

Table 14: In-Process Tablet Specifications

Parameters	Target
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Average Tablet Weight	225 mg
Individual Thickness	4.0 - 4.5 mm (guideline only)
Individual Tablet Hardness	10 kP
Friability (100 drops)	NO MORE THAN 0.6%
Individual Disintegration	NO MORE THAN 15 min

[000234] Charge the blend into the hopper. Set the turret speed to an appropriate speed. Adjust the die fill amount and compression parameters to yield a tablet with a target weight of 225 mg and with a target hardness of 8-12 kP. All waste material should be collected in the tableting waste poly bag. Start timer and compress tablets.

Example 10

290µg Tablet Blending and Compression

[000235] According to the procedure of Example 7, a 290µg dose tablet of linaclotide may be prepared with the formulation of Table 15.

Table 15: Formulation of a 290 µg Linaclotide Tablet:

Component	% w/w	Quantity (g)
Pregranulated Fillers	60.10	3906.5
Linaclotide Base	38.65	2512.3
Magnesium Stearate	1.25	81.3
Total	100	6500

[000236] First, dispense the raw materials of Table 16.

Table 16: Raw materials for preparation of 290µg linaclotide tablet

Raw Material	Required Quantity (g)
Pregranulated Fillers	3906.6
Linaclotide Base	2512.2
Magnesium Stearate	81.2
Total	6500.0

[000237] Sub-blend A: Add the Pregranulated Fillers and Linaclotide base for sub-blend A to a 16 qt v-blender. Close lids and blend for 10 minutes. Pass the Magnesium Stearate for sub-blend A through the 40 mesh screen. Add to the 16 qt v-blender. Close lids and blend for 3 minutes. Tare a poly bag, and discharge the blend into it.

[000238] Sub-blend B: Add the Pregranulated Fillers and Linaclotide Base for sub-blend B to a 16 qt v-blender. Close lids and blend for 10 minutes. Pass the Magnesium Stearate for sub-blend B through the 40 mesh screen. Add to the 16 qt v-blender. Close lids and blend for 3 minutes. Tare a poly bag, and discharge the blend into it.

[000239] Compression: Ensure a Korsch XL 100 tablet press with 0.32" sc toolings is set up with 0.32" round concave tooling (10 stations desired), paddle feeder, and that all other components are properly secured. Manually rotate die table at least one full revolution to ensure proper installation. Visually confirm presence of all dies and punches, and that dies are flush with the die table. Verify that the tablet specifications meet the In-Process Tablet Specifications of Table 17.

Table 17: In-Process Tablet Specifications

Parameters	Target
Average Tablet Weight	225 mg
Individual Thickness	4.0 - 4.5 mm (guideline only)
Individual Tablet Hardness	10 kP
Friability (100 drops)	NO MORE THAN 0.6%
Individual Disintegration	NO MORE THAN 15 min

[000240] Charge the blend into the hopper. Set the turret speed to an appropriate speed. Adjust the die fill amount and compression parameters to yield a tablet with a target weight of 225 mg and with a target hardness of 8-12 kP. All waste material should be collected in the tableting waste poly bag. Start timer and compress tablets.

Example 11

Sub-Coating of Linaclotide 25 µg Tablets

[000241] The 25µg tablets of Example 8 may be sub-coated with an Opadry® II sub-coating. The formula of Table 18 represents the amounts needed to prepare the coating material, which is prepared in excess to account for losses in the coating process.

Table 18: Sub-Coating formula for 25µg tablets

Component	%w/w	g/batch
Opadry II White	20	400
Purified water	80	1600
Total	100	2000

[000242] Dispense 1600g of purified water into a suitably sized container. Dispense 400g of Opadry® II into a suitably sized container. Add Opadry® to the water. Calculate the theoretical amount of solution needed to apply a 4.0% weight gain, with 85% theoretical efficiency. Prepare a poly bag for the waste tablets collected during the coating process. Ensure the Compu-Lab has been set up with the 19 inch pan and plenum assembly. Verify the liquid feed lines are Tygon 17 tubing. Verify the gun assembly is installed in the pan. The spray gun assembly should consist of 1 x ¼ JAU spray gun mounted with a 40100 AB liquid spray nozzle and matching air cap. The gun assembly should be mounted as far as possible from the tablet bed, with the spray angle perpendicular to the top third of the bed. Verify initial tablet weight by weighing 100 uncoated tablets. Calculate the average weight of a single tablet by dividing the weight of the tablets by 100. Test the initial tablet moisture by crushing approximately 1 gram of tablets and running LOD for 10 minutes at 105°C. Adjust the pump so that the liquid flow rate is approximately 10 g/min. Prime the lines past the guns and verify that there is no leaking in the lines or gun. Charge the tablets into the coating pan and begin warming for 20 minutes with an inlet temperature of 50°C and airflow of 350 CFM. Jog occasionally during warm-up.

[000243] Once target bed temperature is reached, begin spraying the coating suspension according to the target process parameters outlined in Table 19 below. Test and report tablet moisture content periodically as desired. Once the theoretical amount of solution has been applied check the tablets for weight gain. When 4% weight gain has been achieved, stop spray and dry tablets for 30 minutes with an inlet temperature of 50°C, reducing pan speed to a minimum or jogging. Discharge the tablets and determine the new weight of the coated tablets.

Table 19: Target Process Parameters

Parameters	Target
Spray rate	10 g/min
Inlet temperature	65°C
Airflow	350 CFM

Atomization air	40 PSI
Pan speed	11 RPM
Exhaust temperature	47°C
Bed Temperature	50°C

Example 12

Sub-Coating of 100 µg Linacotide Tablets

[000244] The 100µg tablets of Example 9 may be sub-coated with an Opadry® II coating. The formula of Table 20 represents the amounts needed to prepare the coating material, which is prepared in excess to account for losses in the coating process.

Table 20: Sub-Coating formula for 100µg tablets

Component	%w/w	g/batch
Opadry II White	20	1000
Purified water	80	4000
Total	100	5000

[000245] Dispense 4000g of purified water into a suitably sized container. Dispense 1000g of Opadry® II into a suitably sized container. Add Opadry® to the water. Calculate the theoretical amount of solution needed to apply a 4.0% weight gain, with 85% theoretical efficiency. Ensure the Compu-Lab has been set up with the 24 inch pan and plenum assembly. Verify the liquid feed lines are Tygon 17 tubing. Verify the gun assembly is installed in the pan. The spray gun assembly should consist of 2 x ¼ JAU spray gun mounted with a 40100 AB liquid spray nozzle and matching air cap. The gun assembly should be mounted as far as possible from the tablet bed, with the spray angle perpendicular to the top third of the bed. Verify initial tablet weight by weighing 100 uncoated tablets. Calculate the average weight of a single tablet by dividing the weight of the tablets by 100. Test the initial tablet moisture by crushing approximately 1 gram of tablets and running LOD for 10 minutes at 105°C. Adjust the pump so that the liquid flow rate is approximately 20 g/min. Prime the lines past the guns and verify that there is no leaking in the lines or gun. Charge the tablets into the coating pan and begin warming for 20 minutes with an inlet temperature of 50°C and airflow of 400 CFM. Jog occasionally during warm-up.

[000246] Once target bed temperature is reached, begin spraying the coating suspension according to the target process parameters outlined in Table 21 below. Test and report tablet moisture content periodically as desired. Once the theoretical amount of solution has been

applied check the tablets for weight gain. When 4% weight gain has been achieved, stop spray and dry tablets for 30 minutes with an inlet temperature of 50°C, reducing pan speed to a minimum or jogging. Discharge the tablets and determine the new weight of the coated tablets.

Table 21: Target Process Parameters

Parameters	Target
Spray rate	20 g/min
Inlet temperature	65°C
Airflow	400 CFM
Atomization air	40 PSI
Pan speed	10 RPM
Exhaust temperature	47°C
Bed Temperature	50°C

Example 13

Sub-Coating of 290 µg Linacotide Tablets

[000247] The 290µg tablets of Example 10 may be sub-coated with an Opadry® II coating. The formula of Table 22 represents the amounts needed to prepare the coating material, which is prepared in excess to account for losses in the coating process.

Table 22: Sub-Coating formula for 100µg tablets

Component	%w/w	g/batch
Opadry II White	20	400
Purified water	80	1600
Total	100	2000

[000248] Dispense 1600g of purified water into a suitably sized container. Dispense 400g of Opadry® II into a suitably sized container. Add Opadry® to the water. Calculate the theoretical amount of solution needed to apply a 4.0% weight gain, with 85% theoretical efficiency. Ensure the Compu-Lab has been set up with the 19 inch pan and plenum assembly. Verify the liquid feed lines are Tygon 17 tubing. Verify the gun assembly is installed in the pan. The spray gun assembly should consist of 1 x ¼ JAU spray gun mounted with a 40100 AB liquid spray nozzle and matching air cap. The gun assembly should be mounted as far as possible from the tablet bed, with the spray angle perpendicular to the top third of the bed. Verify initial tablet weight by weighing 100 uncoated tablets. Calculate the

average weight of a single tablet by dividing the weight of the tablets by 100. Test the initial tablet moisture by crushing approximately 1 gram of tablets and running LOD for 10 minutes at 105°C. Adjust the pump so that the liquid flow rate is approximately 10 g/min. Prime the lines past the guns and verify that there is no leaking in the lines or gun. Charge the tablets into the coating pan and begin warming for 20 minutes with an inlet temperature of 50°C and airflow of 350 CFM. Jog occasionally during warm-up.

[000249] Once target bed temperature is reached, begin spraying the coating suspension according to the target process parameters outlined in Table 23 below. Test and report tablet moisture content periodically as desired. Once the theoretical amount of solution has been applied check the tablets for weight gain. When 4% weight gain has been achieved, stop spray and dry tablets for 30 minutes with an inlet temperature of 50°C, reducing pan speed to a minimum or jogging. Discharge the tablets and determine the new weight of the coated tablets.

Table 23: Target Process Parameters

Parameters	Target
Spray rate	10 g/min
Inlet temperature	65°C
Airflow	350 CFM
Atomization air	40 PSI
Pan speed	1 RPM
Exhaust temperature	47°C
Bed Temperature	50°C

Example 14

Functional or Enteric Coating of Linaclotide Tablets (“DR1”)

[000250] The tablets of previous examples may be prepared with a functional coating. The formulation of Table 24 represents the amounts needed to prepare the coating material, which is prepared in excess to account for losses in the coating process:

Table 24: Formulation for functional coating process

Component	%w/w
Eudragit ® S100	8.25
Eudragit ® L100	1.69
1N NH ₃	6.75
Triethyl Citrate	4.97
Talc	4.97
Purified water	73.37
Total	100

[000251] To prepare the functional coating, dispense the required quantity of Eudragit® S100 and Eudragit® L100 into a suitably sized container. Dispense 2/3 of the required quantity of purified water into a suitably sized container. Begin agitation of water until a vortex is achieved. Add Eudragit® S100 and Eudragit® L100 slowly to the water and mix until the powder is thoroughly wetted and lump or foam formation has dissipated (about 5 minutes).

[000252] Dispense the required quantity of 1N NH₃ into a suitable sized container. Add the 1N NH₃ slowly into the Eudragit® suspension and mix for a minimum of 60 minutes. Dispense the required quantity of Triethyl Citrate into a suitable sized container. Add the Triethyl Citrate into the Eudragit® suspension and mix for a minimum of 60 minutes. Dispense the required quantity of Talc into a suitable sized container. Homogenize the talc in the remaining 1/3 of purified water for 10 minutes (or until homogenous) using a Silverson Homogenizer. Pour the talc suspension into the Eudragit® suspension while mixing. Mix for no longer than 5 minutes. Screen the coating suspension through a #30 mesh screen.

[000253] Calculate the theoretical amount of solution needed to apply a 9.0% weight gain, with 90% theoretical efficiency. Ensure the Compu-Lab has been set up with the 19 inch pan and plenum assembly. Verify the liquid feed lines are Tygon 17 tubing. Verify the gun assembly is installed in the pan. The spray gun assembly should consist of 1 x ¼ JAU spray gun mounted with a 40100 AB liquid spray nozzle and matching air cap. The gun assembly should be mounted as far as possible from the tablet bed, with the spray angle perpendicular to the top third of the bed. Charge the tablets into the coating pan. Warm the tablets with an inlet temperature of 30 degrees. Ensure bed temperature reaches approximately 30 degrees before proceeding to next step. Adjust the pump so that the liquid flow rate is approximately 12 g/min. Prime the lines past the guns and verify that there is no leaking in the lines or gun. Test the starting tablet moisture by crushing approximately 2 gram of tablets and running LOD for 10 minutes at 105°C.

[000254] Once target bed temperature is reached, begin spraying the coating suspension according to the target process parameters outlined in Table 25 below. Test and report tablet moisture content periodically as desired. Once the theoretical amount of solution has been applied check the tablets for weight gain. When 9% weight gain has been achieved, stop spray and dry tablets for 5-10 minutes with an inlet temperature of 40°C, reducing pan speed to a minimum or jogging.

Table 25: Target process parameters for spray coating process

Parameters	Target
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Spray rate	12 – 20 g/min ¹
Inlet temperature	43°C ¹
Airflow	300 CFM
Atomization air	35 PSI
Pan speed	12 RPM
Exhaust temperature	33°C
Bed Temperature	35°C

¹ Adjust as needed to maintain bed temperature.

[000255] Test the final tablet moisture by crushing approximately 2 gram of tablets and running LOD for 10 minutes at 105°C. Moisture should be ≤ the initial tablet moisture. Discharge the tablets and determine the new weight of the coated tablets. Dry tablets for at least 2 hours in a mechanical convection oven with the temperature set to 40°C. Bulk package tablets in foil bag with desiccants and store at 5°C. The coating of this Example 14 is used as a coating for the DR1 delayed release compositions discussed herein.

Example 15

Alternative Functional or Enteric Coating of Linacotide Tablets (“DR2”)

[000256] An alternative coating, may be prepared according to the method of Example 14 but with the formulation of Table 26. The coating of this Example 15 is used as a coating for the DR2 delayed release compositions discussed herein.

Table 26: Formulation for functional coating process

Component	%w/w
Eudragit ® S100	9.94
1N NH ₃	6.75
Triethyl Citrate	4.97
Talc	4.97
Purified water	73.37
Total	100

Example 16

Organic Coating of Linacotide Tablets

[000257] An organic coating may be provided for the linacotide tablets of the examples above. For the coating of 100 µg tablets, the formula of Table 27 was used.

Table 27: Organic coating material formula for linacotide tablets.

Component	%w/w	g/batch
Eudragit S100	2.941	88.2
Eudragit L100	2.941	88.2
Triethyl Citrate	1.177	35.3

Talc	2.941	88.2
Acetone	34.290	1028.7
Isopropanol	51.420	1542.6
Purified Water	4.290	128.7
Total	100	3000

[000258] Dispense the required quantity of purified water into a suitable sized container. Dispense the required quantity of acetone into a suitable sized container. Begin mixing acetone and add the water. Dispense the required quantity of isopropanol into a suitable sized container. Add isopropanol to the water and acetone to create the diluent mixture. Pour approximately half of the diluent mixture into a second container and resume mixing the first half of the diluent mixture. Dispense the required quantity of Eudragit S100 into a suitable sized container. Begin mixing the second half of diluent mixture and add the Eudragit S100. Dispense the required quantity of Eudragit L100 into a suitable sized container.

[000259] Add the Eudragit L100 to the diluent mixture containing the Eudragit S100 and start timer. Stir until the polymers are completely dissolved. Add the triethyl citrate to the first half of the diluent mixture while mixing with a high shear mixer. Dispense the required quantity of talc into a suitable sized container. Add the talc to the first half of the diluent mixture while mixing with a high shear mixer to create the excipient suspension. Start the timer and mix for at least 10 minutes. Record the mixing time of the Eudragit solution. While continuing to mix the Eudragit solution, slowly pour the excipient suspension into the Eudragit solution. Once the coating suspension is uniform pass it through a 35 mesh screen. Resume mixing. Calculate the theoretical amount of solution needed to apply a 8.5% weight gain, with 88% theoretical efficiency.

[000260] Prepare a poly bag for the waste tablets collected during the coating process. Ensure the Compu-Lab has been set up with the 15 inch pan and plenum assembly. Verify the liquid feed lines are solvent resistant 17 tubing. Verify the gun assembly is installed in the pan. The spray gun assembly should consist of 1 x ¼ JAU spray gun mounted with a 40100 AB liquid spray nozzle and matching air cap. The gun assembly should be mounted as far as possible from the tablet bed, with the spray angle perpendicular to the top third of the bed. Adjust the pump so that the liquid flow rate is approximately 28 g/min. Prime the lines past the guns and verify that there is no leaking in the lines or gun. Charge the tablets into the coating pan and begin warming with an inlet temperature of 38°C and airflow of 200 CFM. Jog occasionally during warm-up. Once the product temperature reaches about 27°C, begin

spraying the coating suspension according to the target process parameters outlined below. When 8.5% weight gain has been achieved, stop spray and dry tablets for 10 minutes with an inlet temperature of 40°C, reducing pan speed to a minimum or jogging.

Parameters	Target
Spray rate	27-30 g/min
Inlet temperature	30-40°C
Airflow	200 CFM
Atomization air	17 PSI
Pan speed	13 RPM
Exhaust temperature	25-28°C
Bed Temperature	26-27°C

[000261] Place tablets onto trays and dry for at least 24 hours in a mechanical convection oven with the temperature set to 45°C.

Example 17

Alternative Linacotide Tablet Preparation

[000262] Delayed release tablets were prepared by first preparing the following core tablet components: a placebo base, a linacotide 1000 µg/225 mg base, and pre-granulated fillers.

Granulation manufacturing process:

[000263] The tablet components were prepared essentially as described above in Example 7, but with slight modifications as described below. The placebo base as described in Table 28 was used to provide core tablets with the components listed in Table 28. A final pH of 2.25 was used for placebo and active base granulations.

Table 28: Components for various tablet strengths

Strength	Placebo	25 µg	30µg	50 µg	75 µg	100 µg	150 µg	290 µg	300µg
Placebo base (%)	30	27.5	27	25	22.5	20	15	1	0
Linacotide base (i.e., 1000 ug/225 mg base (%))	0	2.5	3	5	7.5	10	15	29	30
Pregranulated fillers (%)	68.75	68.75	68.75	68.75	68.75	68.75	68.75	68.75	68.75
Magnesium Stearate (%)	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25	1.25

Total (%)	100	100	100	100	100	100	100	100	100
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Placebo Base Preparation:

[000264] Table 29 represents the formulation for the placebo base granulation:

Table 29: Formulation of the placebo base granulation

Component	%w/w	Quantity (g)
L-Histidine	4.97	248.5
Calcium Chloride Dihydrate	2.35	117.5
polyvinyl alcohol	3.96	198
microcrystalline cellulose	88.72	4436
Hydrochloric acid, pure, fuming, 37% solution in water		
Treated water		
Total	100	5000

[000265] The placebo base preparation was prepared by first dispensing the raw materials of Table 30.

Table 30: Raw materials for placebo base preparation

Component	Quantity (g)
L-Histidine	248.4
Calcium Chloride Dihydrate	117.7
polyvinyl alcohol	198
microcrystalline cellulose	4435.9

[000266] The mix container is tared and 2200 ± 5.0 g of treated water (rather than 2682.1 ± 5.0 g) is added into the container.

Linacotide Base Preparation (1000 μ g/225mg):

[000267] Table 31 represents the formulation for the 1000 μ g/225mg base granulation:

Table 31: Formulation for the 1000 μ g/225mg base granulation

Component	% w/w	Quantity (g)
Linacotide	0.444	23
L-Histidine	4.97	248.4
Calcium Chloride Dihydrate	2.35	117.7
polyvinyl alcohol	3.96	198
microcrystalline cellulose	88.27	4413.7

A balance of 0.02% represents Linacotide impurities.

[000268] The 1000 μ g/225mg base granulation may be prepared by first dispensing the raw materials of Table 32.

Table 32: Raw materials of the linacotide base granulation

Raw Material	Required Quantity (g)
Linacotide	23
microcrystalline cellulose	4413.7
Granulation solution	

Pregranulated Filler Preparation:

[000269] Table 33 represents the formulation for the pregranulated fillers.

Table 33: Formulation of the fillers granulation

Component	% w/w	Qty per subplot (g)
microcrystalline cellulose	14.6	1018
croscarmellose sodium	2.9	203.6
mannitol	79.1	5533.2
polyvinyl alcohol	3.5	245
Treated water	-----	-----
Total	100.0	7,000

[000270] The fillers preparation are prepared by first dispensing the raw materials of Table 34.

Table 34: Raw materials for preparation of fillers granulation

Raw Material	Required Quantity (g)
polyvinyl alcohol	245
microcrystalline cellulose	1018
croscarmellose sodium	203.6
mannitol	5533.2
Total	---

Example 18

Alternative 225 mg Linacotide Tablet Formulations

[000271] Delayed release 225 mg tablets can be prepared with the proportions of excipients as shown in Table 34 and Table 35. The 225 mg tablets of this Example 18 can be

prepared at dosage strengths of 30 µg, 100 µg, or 300 µg. Two example embodiments, A and B, of the proportions of excipients are provided for each dosage strength. Embodiment A (at each dosage) in Table 34 corresponds to the DR1 formulation in Example 20; embodiment A (at each dosage) in Table 35 corresponds to the DR2 formulation in Example 20. The tablets include a tablet core, a barrier coat, a functional coat, and, in some cases, an aesthetic coat.

[000272] Table 34: Alternative 225mg tablet formulation (A=DR1)

Component	%w/w					
	30 µg strength		100 µg strength		300 µg strength	
	A	B	A	B	A	B
Tablet Core						
linacotide ^a	0.013	0.013	0.044	0.044	0.132	0.132
L-Histidine	0.5	1.49	0.5	1.49	0.9	1.49
Calcium Chloride Dihydrate	0.2	0.71	0.2	0.71	0.4	0.71
Polyvinyl Alcohol	3.3	3.59	3.3	3.59	2.7	3.59
MCC PH 200 ^a	34.3	41.73	34.3	41.70	49.3	41.61
Mannitol 100SD	56.5	46.22	56.5	46.22	42.1	46.22
Crosscarmellose sodium	4.00	5.00	4.00	5.00	3.0	5.00
Magnesium Stearate	1.25	1.25	1.25	1.25	1.25	1.25
Purified Water	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b
Total (Tablet Core)	100.0	100.0	100.0	100.0	100.0	100.0
Barrier Coat Suspension						
Opadry II White 85F18422	4 ^c	4 ^c	4 ^c	4 ^c	4 ^c	4 ^c
Purified Water	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b
Functional Coat Suspension						
Eudragit S100 ^d	3.9	3.9	3.9	3.9	3.9	3.9
Eudragit L100 ^d	0.8	0.8	0.8	0.8	0.8	0.8
Triethyl citrate	2.3	2.3	2.3	2.3	2.3	2.3
Talc	2.3	2.3	2.3	2.3	2.3	2.3
Purified Water	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b
Aesthetic Coat Suspension						
Opadry II TBD ^{c,e}	--	4	--	4	--	4
Purified Water	--	-- ^b	--	-- ^b	--	-- ^b

a The actual quantity of linacotide drug substance is adjusted based on the purity of the drug substance with a concomitant adjustment of microcrystalline cellulose

b Purified water is removed during drug product processing

c Represents 4% weight gain on previous step

d Represents 9.5% total Eudragit weight gain on previous step

e Final Opadry will be either Blue 85F99031, Yellow 85F120017, or Orange 85F130136

[000273] Table 35: Alternative 225mg tablet formulation (A=DR2)

Component	%w/w					
	30 µg strength		100 µg strength		300 µg strength	
	A	B	A	B	A	B
Tablet Core						
linacotide ^a	0.013	0.013	0.044	0.044	0.132	0.132
L-Histidine	0.5	1.49	0.5	1.49	0.9	1.49
Calcium Chloride Dihydrate	0.2	0.71	0.2	0.71	0.4	0.71
Polyvinyl Alcohol	3.3	1.88	3.3	1.88	2.7	1.88
MCC PH 200 ^a	34.3	41.73	34.3	41.70	49.3	41.61
Mannitol 100SD	56.5	47.94	56.5	47.94	42.1	47.94
Crosscarmellose sodium	4.00	5.00	4.00	5.00	3.0	5.00
Magnesium Stearate	1.25	1.25	1.25	1.25	1.25	1.25
Purified Water	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b
Total (Tablet Core)	100.0	100.0	100.0	100.0	100.0	100.0
Barrier Coat Suspension						
Opadry II White 85F18422	4 ^c	4 ^c	4 ^c	4 ^c	4 ^c	4 ^c
Purified Water	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b
Functional Coat Suspension						
Eudragit S100 ^d	4.7	4.7	4.7	4.7	4.7	4.7
Triethyl citrate	2.3	2.3	2.3	2.3	2.3	2.3
Talc	2.3	2.3	2.3	2.3	2.3	2.3
Purified Water	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b	-- ^b
Aesthetic Coat Suspension						
Opadry II TBD ^{c,e}	--	4	--	4	--	4
Purified Water	--	-- ^b	--	-- ^b	--	-- ^b

a The actual quantity of linacotide drug substance is adjusted based on the purity of the drug substance with a concomitant adjustment of microcrystalline cellulose

b Purified water is removed during drug product processing

c Represents 4% weight gain on previous step

d Represents 9.5% total weight gain on previous step

e Final Opadry may be either Blue 85F99031, Yellow 85F120017, or Orange 85F130136

Example 19

Alternative 450 mg Linacotide Tablet Formulations

[000274] Delayed release 450 mg tablets can be prepared with the proportions of excipients as shown in Table 36 and Table 37. The 450 mg tablets of this Example 19 can be prepared at dosage strengths of 300 or 600 µg. The tablets include a tablet core, a barrier coat, a functional coat, and an aesthetic coat.

[000275] Table 36: Alternative 450 mg tablet formulation

Component	%w/w	
	300 µg	600 µg
Tablet Core		
linacotide ^a	0.066	0.132
L-Histidine	1.49	1.49
Calcium Chloride Dihydrate	0.71	0.71
Polyvinyl Alcohol	3.59	3.59
MCC PH 200 ^a	41.68	41.61
Mannitol 100SD	46.22	46.22
Crosscarmellose sodium	5.00	5.00
Magnesium Stearate	1.25	1.25
Purified Water	-- ^b	-- ^b
Total (Tablet Core)	100.0	100.0
Barrier Coat Suspension		
Opadry II White 85F18422	3	3
Purified Water	-- ^b	-- ^b
Functional Coat Suspension		
Eudragit S100 ^d	2.9	2.9
Eudragit L100 ^d	0.5	0.5
Triethyl citrate	1.8	1.8
Talc	1.8	1.8
Purified Water	-- ^b	-- ^b
Aesthetic Coat Suspension		
Opadry II TBD ^{c,e}	3	3
Purified Water	-- ^b	-- ^b

a The actual quantity of linacotide drug substance is adjusted based on the purity of the drug substance with a concomitant adjustment of microcrystalline cellulose

b Purified water is removed during drug product processing

c Represents 3% weight gain on previous step

d Represents 7.1% total Eudragit weight gain on previous step

e Final Opadry may be either Blue 85F99031, Yellow 85F120017, or Orange 85F130136

[000276] Table 37: Alternative 450 mg tablet formulation

Component	%w/w

	300 µg	600 µg
Tablet Core		
linaclotide ^a	0.066	0.132
L-Histidine	1.49	1.49
Calcium Chloride Dihydrate	0.71	0.71
Polyvinyl Alcohol	1.88	1.88
MCC PH 200 ^a	41.68	41.61
Mannitol 100SD	47.94	47.94
Crosscarmellose sodium	5.00	5.00
Magnesium Stearate	1.25	1.25
Purified Water	-- ^b	-- ^b
Total (Tablet Core)	100.0	100.0
Barrier Coat Suspension		
Opadry II White 85F18422	3 ^c	3 ^c
Purified Water	-- ^b	-- ^b
Functional Coat Suspension		
Eudragit S100 ^d	3.5	3.5
Triethyl citrate	1.8	1.8
Talc	1.8	1.8
Purified Water	-- ^b	-- ^b
Aesthetic Coat Suspension		
Opadry II TBD ^{c,e}	3	3
Purified Water	-- ^b	-- ^b

a The actual quantity of linaclotide drug substance is adjusted based on the purity of the drug substance with a concomitant adjustment of microcrystalline cellulose

b Purified water is removed during drug product processing

c Represents 3% weight gain on previous step

d Represents 7.1% total weight gain on previous step

e Final Opadry will be either Blue 85F99031, Yellow 85F120017, or Orange 85F130136

Example 20

Administration of Linaclotide for the Treatment of Irritable Bowel Syndrome with Constipation (IBS-c)

[000277] Delayed release linaclotide tablets were administered in a Phase 2b, randomized, double-blind, double-dummy, placebo-controlled, parallel-group, dose-range-finding study of two delayed release formulations (“DR1” and “DR2”) of linaclotide administered orally for 12 weeks to patients with IBS-c (the “Trial”).

[000278] Trial methodology:

[000279] The Trial was a multi-center, multiple-dose study consisting of 3 distinct periods: a screening period, a pretreatment period and a treatment period (see Fig. 1). The

study enrolled patients having IBS-c diagnosed using Rome III criteria. Eligible patients were randomized in equal proportions to one of 8 treatments: 30 µg, 100 µg, or 300 µg of the linaclotide DR1 formulation; 30 µg, 100 µg, or 300 µg of the linaclotide DR2 formulation; 290 µg of the immediate release (IR) formulation of linaclotide; or placebo. Linaclotide IR was included as a positive control to compare safety and efficacy. The DR1 delayed release composition was designed to release the linaclotide core tablet in the ileal region. The DR2 delayed release composition was designed to release the linaclotide core tablet within the late ileum to the ascending colon (i.e., at or near the ileocecal junction).

[000280] Pre-treatment period:

[000281] After a screening period (to determine patient eligibility), a pre-treatment period began. The pre-treatment period was defined as the 14 to 21 calendar days immediately before starting the treatment period. During the pretreatment period, patients provided information regarding daily bowel-movement-related symptom severity, daily abdominal symptom-severity, weekly constipation severity, weekly IBS symptom severity, weekly adequate relief and use of per-protocol rescue medicine or other laxatives, suppositories or enemas on an event-driven basis. Patients who satisfied all of the entry criteria entered the treatment period. The patient information from the pre-treatment period was used for baseline characteristic data as compiled in Table 38. Pain, Bloating and Discomfort were on an 11-point NRS (0=None; 10=Worse possible). Straining was on a 5-point score (1=None at all; 5=Extreme amount).

Table 38: Patient Baseline Characteristics

	Placebo (N=66)	IR 290 ug (N=66)	DR 1			DR 2		
			30 ug (N=67)	100 ug (N=67)	300 ug (N=67)	30 ug (N=67)	100 ug (N=66)	300 ug (N=66)
Pain ¹	6.4	6.2	6.2	6.2	6.4	6.1	6.5	6.1
CSBM/Week	0.3	0.3	0.4	0.3	0.3	0.4	0.3	0.3
SBM/Week	1.7	1.8	1.7	1.6	1.7	1.7	1.6	1.7
BSFS	2.3	2.2	2.3	2.4	2.2	2.3	2.4	2.2
Symptom Composite	6.6	6.3	6.4	6.4	6.7	6.4	6.4	6.7
Bloating ¹	6.7	6.4	6.5	6.9	6.6	6.7	6.5	7
Discomfort ¹	6.6	6.3	6.4	6.7	6.4	6.5	6.3	6.7
Straining ²	3.4	3.5	3.4	3.6	3.6	3.4	3.6	3.6

[000282] Treatment period:

[000283] The treatment period began with treatment assignment (randomization) and lasted for 12 weeks. Patients were randomly assigned to one of 8 treatments with linaclotide: 30 µg, 100 µg, or 300 µg of the linaclotide DR1 formulation; 30 µg, 100 µg, or 300 µg of the

linaclotide DR2 formulation; 290 µg of the IR formulation; or placebo. Patients were instructed to take the study drug once daily in the morning at least 30 minutes before breakfast. Patients continued to provide assessments regarding daily bowel-movement-related symptom severity, daily abdominal symptom-severity, weekly constipation severity, weekly IBS symptom severity, weekly adequate relief, and use of per-protocol rescue medicine or other laxatives, suppositories or enemas on an event-driven basis.

[000284] In the Trial, the daily and weekly assessment data for patients in each treatment group was compiled to give the following primary and secondary treatment efficacy parameters, which have been defined by U.S. and European regulatory agencies for the study of the efficacy of IBS-c.

[000285] Primary efficacy parameters:

[000286] The weekly change from baseline in abdominal pain;

[000287] The weekly change from baseline in CSBM frequency; and

[000288] 6/12 week abdominal pain and constipation (APC) +1 responder. A patient was classified as a 6/12 week abdominal pain and constipation (APC) +1 responder if they were a weekly APC +1 responder for ≥ 6 out of the 12 weeks of the treatment period. A weekly APC +1 responder was defined as a patient who meets both the criteria to be a weekly CSBM +1 responder and a weekly abdominal pain responder in a given week.

[000289] Secondary efficacy parameters:

[000290] 6/12 week CSBM +1 responder; A patient was classified as a 6/12 week CSBM +1 responder if they were a weekly CSBM +1 responder for ≥ 6 out of the 12 weeks of the treatment period. A weekly CSBM +1 responder was defined as a patient who had an increase from baseline of at least 1 in the CSBM weekly rate for a given week.

[000291] 6/12 week abdominal pain responder; A patient was classified as a 6/12 week abdominal pain responder if they were a weekly abdominal pain responder for ≥ 6 out of the 12 weeks of the treatment period. A weekly abdominal pain responder was defined as a patient who had a decrease from baseline of at least 30% in the mean abdominal pain score for a given week.

[000292] Change from baseline in 12-week (overall treatment period) abdominal pain;

[000293] Change from baseline in 12-week (overall treatment period) CSBM frequency rate;

[000294] Change from baseline in 12-week (overall treatment period) SBM frequency rate;

[000295] Change from baseline in 12-week (overall treatment period) stool consistency (BSFS);

- [000296] Change from baseline in 12-week (overall treatment period) straining;
- [000297] Change from baseline in 12-week (overall treatment period) abdominal discomfort;
- [000298] Change from baseline in 12-week (overall treatment period) abdominal bloating;
- [000299] Change from baseline in 12-week (overall treatment period) constipation severity;
- [000300] Change from baseline in 12-week (overall treatment period) abdominal symptom score; and
- [000301] Change from baseline in 12-week (overall treatment period) IBS symptom severity;
- [000302] Change from baseline in 12-week (overall treatment period) percent of days with use of per-protocol rescue medicine;
- [000303] Treatment satisfaction; and
- [000304] Assessment of adequate relief.
- [000305] Results of Trial:
- [000306] The results of the efficacy parameters for the Trial are shown in Figures 2-13, 17 and 21. The data in these figures clearly indicate clinically meaningful and statistically significant improvement was achieved for IBS-c patients treated with delayed release linaclotide compositions compared to the placebo group and the immediate release linaclotide group. The improvement was shown across all three primary efficacy parameters, the secondary efficacy parameters, and the occurrence of adverse events.
- [000307] The results of the Trial demonstrate that administration of the DR1 composition has efficacy across the primary and secondary efficacy parameters. The results of the Trial also demonstrate that administration of the DR2 compositions had no effect on constipation as shown by CSBM and BSFS data (e.g., Figures 5 and 11); however administration of the DR2 compositions did improve symptoms of abdominal pain in the patients (with very low incidence of adverse events), suggesting that the DR2 composition is useful for reducing abdominal symptoms (pain, discomfort, bloating) in non-constipated patients.
- [000308] Figure 2 shows the 6/12 Week APC +1 parameter results. The 300 µg DR1 composition showed improvement over the comparable IR composition (38.8% vs. 31.8%). Other DR1 and DR2 compositions showed improvement over placebo.
- [000309] Figure 3 shows the weekly change from baseline in SBM frequency.

[000310] Figure 4 shows the weekly change from baseline in abdominal pain for each week over the 12 week treatment period. Again the 300 µg DR1 composition outperformed the comparable IR composition and other DR compositions outperformed placebo. In addition, the 30 µg DR2 composition showed a change in abdominal pain levels comparable to the IR composition.

[000311] Figures 5-13 show the trial results for secondary efficacy parameters, demonstrating the overall improvement in patient outcomes for the delayed release compositions. In past studies, the Bristol Stool Form Scale has been found to be one of the most reliable and consistent endpoints as a measure of efficacy in IBS-c trials. As shown in Figure 11, DR1 appears to show dose dependent improvement in BSFS scores.

[000312] Figure 17 shows selected efficacy parameter results with comparison between the 300 µg DR1 composition, the 290 µg IR composition from the Trial, placebo, and the 290 µg IR composition from the Phase III trial for Linzess® and the placebo from the Linzess® Phase III trial. The DR1 composition results compare favorably to the results from the IR composition of the Linzess® Phase III trial.

[000313] Figure 21 is a plot of the percent change from baseline in weekly abdominal pain at week 12 for the DR1 composition, the IR composition and placebo.

[000314] Earlier clinical studies tested immediate release doses of linaclotide from 72 mcg to 579 mcg and tested for improvement in abdominal pain. No differences were observed between the 290 mcg and 579 mcg doses in weekly abdominal pain scores. In light of this, the superior pain relief in the 300 mcg dose of DR1 (referred to as CR1 in Fig. 21) is surprising and unexpected.

[000315] Adverse Events:

[000316] The occurrence of adverse events was also monitored during the Trial. A summary of treatment-emergent adverse events (TEAEs) for the Trial are provided in Table 39. SAE refers to a serious adverse event. ADO refers to diarrhea leading to drop out (i.e., discontinuation of treatment).

Table 39: Summary of adverse events

	DR 1					DR 2		
	Placebo (N=66)	IR 290 ug (N=66)	30 ug (N=67)	100 ug (N=67)	300 ug (N=67)	30 ug (N=67)	100 ug (N=66)	300 ug (N=66)
Number of Patients with TEAE	20 (30.3)	27 (40.9)	23 (34.3)	23 (34.3)	28 (41.8)	15 (22.4)	15 (22.7)	20 (30.3)
Number of Patients Who Died	0	0	0	0	0	0	0	0
Number of Patients with SAE	1 (1.5)	1 (1.5)	0	0	0	0	0	0
Number of Patients with ADO	0	4 (6.1)	1 (1.5)	5 (7.5)	3 (4.5)	0	1 (1.5)	0

[000317] Additional data on adverse events are shown in Figures 14-16, 18 and 19. Figures 14-16 shows that all tested dosages of the DR1 and DR2 delayed release compositions had lower rates of diarrhea (overall and ADO) compared to the IR composition group, including lower rates of moderate to severe diarrhea (Fig. 16). Figures 18 and 19 show the diarrhea rates with comparison to the Linzess® Phase III Trial, also showing improvement in adverse diarrhea events for the 300 µg DR1 composition compared to the Linzess® Phase III results.

Example 19

Food Effect Study

[000318] A food effect study with the delayed release compositions of the present invention was undertaken to determine the effect of taking the delayed release compositions on a full stomach (with food) compared to an empty stomach (after fasting). Patients were separated into two groups. Patients in group 1 were given a 300 µg dose of DR1 or DR2 delayed release composition of linaclotide under fed conditions. Specifically, the patients in group 1 were given the composition 30 minutes after the start of a high-fat, high-calorie breakfast with 8 ounces of water. Patients in group 2 were given the same composition (DR1 or DR2) but with no food under fasted conditions. Specifically, the patients in group 2 were given the composition after an overnight fast of at least 10 hours with approximately 8 ounces of water. In both groups, no food was consumed for 4 hours after taking the medication. After a seven day washout period, the patients in group 1 were then given the composition under fasted conditions and the patients in group 2 were given the compositions under fed conditions.

[000319] The results of the study demonstrated that administration to patients of the DR1 composition under fasted conditions had no occurrences of diarrhea. Some patients who took the DR1 composition under fed conditions had occurrences of diarrhea. The patients who took the DR2 composition had no occurrences of diarrhea under fed or fasted conditions. These results are in contrast to earlier studies in healthy subjects using the immediate release dosage form of linaclotide, where administration of 290 µg dose resulted in diarrhea rates of 100% and 94.7% for the fed and fasted subjects, respectively. Therefore, the DR1 and DR2 formulations, which are designed to release in the lower GI as compared with the upper GI release in the immediate release formulation, significantly reduces the incidence of diarrhea to the extent where, in the case of the DR1 formulation, diarrhea is only observed in fed subjects, and in DR2, no diarrhea was observed in either subject group. Therefore, the site of drug release plays a dramatic role in modulating adverse events such as diarrhea.

OTHER EMBODIMENTS

[000320] The present invention is not to be limited in scope by the specific embodiments described herein. Indeed, various modifications of the invention in addition to those described herein will become apparent to those skilled in the art from the foregoing description and the accompanying figures. Such modifications are intended to fall within the scope of the appended claims. It is further to be understood that all values are approximate, and are provided for description.

[000321] All patents, patent applications, publications, product descriptions, and protocols are cited throughout this application, the disclosures of which are incorporated herein by reference in their entireties for all purposes.

CLAIMS

1. A method of reducing the incidence or severity of an adverse event associated with administration of linaclotide, comprising orally administering to a subject a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet comprises an enteric coating comprising a pH-sensitive polymer that releases linaclotide in a lower GI of the subject when compared to administration of an equal dose of an immediate release dosage form of linaclotide, and the delayed-release pharmaceutical tablet composition further comprises a therapeutically effective amount of linaclotide to treat constipation and/or pain in the subject.
2. A method of reducing the incidence or severity of an adverse event associated with administration of linaclotide in a subject, comprising orally administering a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet comprises an enteric coating comprising a pH-sensitive polymer, and the delayed-release pharmaceutical tablet composition releases a therapeutically effective amount of linaclotide to treat constipation and/or pain in the subject.
3. The method of claims 1 or 2, wherein the reduction in the incidence or severity of the adverse event is a reduction in the incidence or severity of the adverse event compared to administration of an equal dose of an immediate release dosage form of linaclotide.
4. The method of any of the preceding claims, wherein the adverse event is diarrhea.
5. The method of any of the preceding claims, wherein the incidence of the adverse event is reduced.
6. The method of any of the preceding claims, wherein the severity of the adverse event is reduced.
7. A method of reducing intestinal fluid secretion-promoting effects of linaclotide, comprising orally administering to a subject a delayed-release pharmaceutical tablet composition comprising linaclotide and the tablet further comprises an enteric coating comprising a pH-sensitive polymer that releases linaclotide in a lower GI of the subject.

8. A method of treating visceral or abdominal pain in a non-constipated subject, comprising orally administering a delayed-release pharmaceutical tablet composition comprising linaclotide, wherein the tablet further comprises an enteric coating and a pH sensitive polymer that releases less than a therapeutically effective amount of linaclotide in a stomach of the subject.
9. A method of treating irritable bowel syndrome with constipation (IBS-c) comprising orally administering to a patient in need thereof, a delayed release pharmaceutical tablet composition comprising a therapeutically effective amount of linaclotide.
10. A method of treating abdominal pain comprising orally administering to a patient in need thereof, a delayed release pharmaceutical tablet composition comprising a therapeutically effective amount of linaclotide.
11. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition comprises a therapeutically effective amount of linaclotide to reduce, prevent or relieve pain or constipation in the subject.
12. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition comprises a therapeutically effective amount of linaclotide to reduce, prevent or relieve pain in the subject, but does not affect bowel habit.
13. The method of claim 12, wherein the bowel habit is selected from CSBM rate, SBM rate, or stool consistency.
14. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition provides less than an amount of linaclotide effective to substantially affect bowel habit.
15. The method of any of the preceding claims, wherein the subject is diagnosed with irritable bowel syndrome with constipation (IBS-c).
16. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition comprising linaclotide releases less than 50%, less than 40%, less than 25% or less than 10% of the linaclotide in the stomach.

17. The method of any of the preceding claims, wherein the linaclotide is present in the delayed-release pharmaceutical tablet composition in an amount between 30 μg to 1,000 μg .
18. The method of claim 17, wherein the linaclotide is present in an amount of about 30 μg , about 100 μg , about 300 μg , about 500 μg , about 600 μg , or about 1,000 μg or wherein the linaclotide is present in an amount of 100 μg , 300 μg , 500 μg , 600 μg , or 1,000 μg .
19. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition further comprises between 0% - 2% per weight of an amino acid selected from the group consisting of alanine, arginine, asparagine, aspartic acid, cysteine, glutamic acid, glutamine, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, proline, serine, threonine, tryptophan, tyrosine, and valine, or any mixture thereof.
20. The method any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition further comprises between 0% - 2% or between 0.5% - 1.5% per weight of histidine.
21. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition further comprises between 0% - 3% per weight of a cation salt selected from the group consisting of calcium, potassium, magnesium, zinc, aluminum, manganese, chromium, cobalt, nickel, barium, and sodium, or any combination or mixture thereof.
22. The method of claim 21, wherein the delayed-release pharmaceutical tablet composition further comprises between 0% - 3% per weight of a calcium salt.
23. The method of claim 22, wherein the delayed-release pharmaceutical tablet composition further comprises between 0% - 2% or between 0.2% - 0.8% per weight of calcium chloride dehydrate.
24. The method of any of the preceding claims, wherein the delayed-release pharmaceutical tablet composition further comprises between 0% - 5%, between 1% - 3%, or between 1% - 1.88% per weight of polyvinyl alcohol (PVA).

25. The method of any of the preceding claims, wherein the pH-sensitive polymer has a dissolution pH of at least 6.0, at least 6.5, or at least 7.0
26. The method of any of the preceding claims, wherein the pH-sensitive polymer comprises a methyl acrylate-methacrylic acid copolymer (e.g., Eudragit®).
27. The method of any of the preceding claims, wherein the pH-sensitive polymer comprises Eudragit S100.
28. The method of any of the preceding claims, wherein the pH-sensitive polymer comprises Eudragit L100.
29. The method of any of the preceding claims, wherein the pH-sensitive polymer consists essentially of Eudragit S100.
30. The method of any of the preceding claims, wherein the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100.
31. The method of claim 30, wherein the pH-sensitive polymer comprises a mixture of Eudragit S100 and Eudragit L100 at a ratio of between 1:1 and 6:1 (S100:L100), at a ratio of between 4.5:1 and 5.5:1 (S100:L100), or at a ratio of 4.875:1 (S100:L100) by weight.
32. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition comprises an enteric coated tablet.
33. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition comprises:
Ca²⁺;
histidine; and
polyvinyl alcohol (PVA).
34. The method of any of the preceding claims, further comprising a protective polymer film or subcoating.
35. The method of claim 34, wherein the subcoating comprises Opadry II®.

36. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition is administered once daily.
37. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition is administered once daily in the morning.
38. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition is administered once daily in the morning at least 30 minutes before breakfast.
39. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition is administered after the patient has fasted for at least 2 hours.
40. The method of any of the preceding claims, wherein the delayed release pharmaceutical tablet composition is administered for at least 12 weeks.
41. The method of any of the preceding claims, wherein the administering increases the number of complete spontaneous bowel movements (CSBMs) by the patient.
42. The method of any of the preceding claims, wherein the administering decreases abdominal pain in the patient.
43. The method of any of the preceding claims, wherein the administering increases the number of complete spontaneous bowel movements (CSBMs) by the patient from baseline by at least one per week during at least 6 out of 12 weeks.
44. The method of any of the preceding claims, wherein the administering decreases the abdominal pain in the patient from baseline by at least 30% during at least 6 out of 12 weeks.
45. The method of any of the preceding claims, wherein the administering increases the number of complete spontaneous bowel movements (CSBMs) by the patient from baseline by at least one per week during at least 6 out of 12 weeks; and wherein the administering

decreases the abdominal pain in the patient from baseline by at least 30% during at least 6 out of 12 weeks.

46. The method of any of the preceding claims, wherein the administering improves one or more of the following: abdominal pain, CSBM frequency rate, SBM frequency rate, stool consistency, straining, abdominal discomfort, abdominal bloating, abdominal symptom score, constipation severity, IBS symptom severity, days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

47. The method of claim 46, wherein the administering improves two or more of the following: abdominal pain, CSBM frequency rate, SBM frequency rate, stool consistency, straining, abdominal discomfort, abdominal bloating, abdominal symptom score, constipation severity, IBS symptom severity, days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

48. The method of any of the preceding claims, wherein the administering results in improvement in one or more of the following efficacy parameters: weekly change from baseline in abdominal pain, weekly change from baseline in CSBM frequency, 6/12 week APC+1 responder, 6/12 week CSBM +1 responder, 9/12 week CSBM +1 responder, 6/12 week abdominal pain responder, 9/12 week abdominal pain responder, weekly CSBM +1 responder, weekly abdominal pain responder, weekly APC +1 responder, change from baseline in 12-week abdominal pain, change from baseline in 12-week CSBM frequency rate, change from baseline in 12-week SBM frequency rate, change from baseline in 12-week stool consistency, change from baseline in 12-week straining, change from baseline in 12-week abdominal discomfort, change from baseline in 12-week abdominal bloating, change from baseline in 12-week abdominal symptom score, change from baseline in 12-week constipation severity, change from baseline in 12-week IBS symptom severity, change from baseline in 12-week percent of days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

49. The method of claim 48, wherein the administering results in improvement in two or more of the following efficacy parameters: weekly change from baseline in abdominal pain, weekly change from baseline in CSBM frequency, 6/12 week APC+1 responder, 6/12 week CSBM +1 responder, 9/12 week CSBM +1 responder, 6/12 week abdominal pain responder, 9/12 week abdominal pain responder, weekly CSBM +1 responder, weekly abdominal pain

responder, weekly APC +1 responder, change from baseline in 12-week abdominal pain, change from baseline in 12-week CSBM frequency rate, change from baseline in 12-week SBM frequency rate, change from baseline in 12-week stool consistency, change from baseline in 12-week straining, change from baseline in 12-week abdominal discomfort, change from baseline in 12-week abdominal bloating, change from baseline in 12-week abdominal symptom score, change from baseline in 12-week constipation severity, change from baseline in 12-week IBS symptom severity, change from baseline in 12-week percent of days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

50. The method of any of the preceding claims, wherein the administering improves, as compared to treatment with an immediate release composition of linaclotide, one or more of the following: abdominal pain, CSBM frequency rate, SBM frequency rate, stool consistency, straining, abdominal discomfort, abdominal bloating, abdominal symptom score, constipation severity, IBS symptom severity, days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

51. The method of any of the preceding claims, wherein the administering results in improvement, as compared to treatment with an immediate release composition of linaclotide, in one or more of the following efficacy parameters: weekly change from baseline in abdominal pain, weekly change from baseline in CSBM frequency, 6/12 week AP+1 responder, 6/12 week CSBM +1 responder, 9/12 week CSBM +1 responder, 6/12 week abdominal pain responder, 9/12 week abdominal pain responder, weekly CSBM +1 responder, weekly abdominal pain responder, weekly APC +1 responder, change from baseline in 12-week abdominal pain, change from baseline in 12-week CSBM frequency rate, change from baseline in 12-week SBM frequency rate, change from baseline in 12-week stool consistency, change from baseline in 12-week straining, change from baseline in 12-week abdominal discomfort, change from baseline in 12-week abdominal bloating, change from baseline in 12-week abdominal symptom score, change from baseline in 12-week constipation severity, change from baseline in 12-week IBS symptom severity, change from baseline in 12-week percent of days with use of per-protocol rescue medicine, treatment satisfaction, and assessment of adequate relief.

52. The method of any of the preceding claims, wherein the administering results in a decrease in the risk of adverse events in the patient compared to treatment with an immediate release composition of linaclotide.

53. The method of claim 52, wherein the administering results in a decrease in the risk of diarrhea in the patient compared to treatment with an immediate release composition of linaclotide.

54. The method of claim 52, wherein the administering results in a decrease of at least 20% in the risk of diarrhea in the patient compared to treatment with an immediate release composition of linaclotide.

55. The method of claim 52, wherein the administering results in a decrease in the risk of severe diarrhea in the patient compared to treatment with an immediate release composition of linaclotide.

56. A method of treating or relieving pain comprising administering to a patient in need thereof a therapeutically effective amount of a delayed-release pharmaceutical tablet composition of any of the preceding claims.

57. The method of claim 56, wherein the pain is selected from visceral pain; diverticulitis pain; pelvic pain; abdominal pain; or pain associated with gastrointestinal disorders, venereal diseases, bladder pain syndrome, or interstitial cystitis.

58. The method of claim 56, wherein the pain is selected from general abdominal pain, diverticular disease, pain associated with irritable bowel syndrome (IBS), chronic or acute radiation proctopathy (also referred to as radiation proctitis), rectal pain, chronic proctalgia, proctalgia fugax, anal pain, chronic anal fissure, post-operative anal pain, overactive bladder syndrome, stress incontinence, interstitial cystitis, bladder pain syndrome, pain associated with cancer, pain associated with gastrointestinal tract neoplasms, general pelvic pain, endometriosis, orchialgia, chronic prostatitis, prostatodynia, vulvodynia, urethral syndrome, penile pain, perianal pain, and pain associated with ulcerative colitis, ulcerative proctitis, or Crohn's disease.

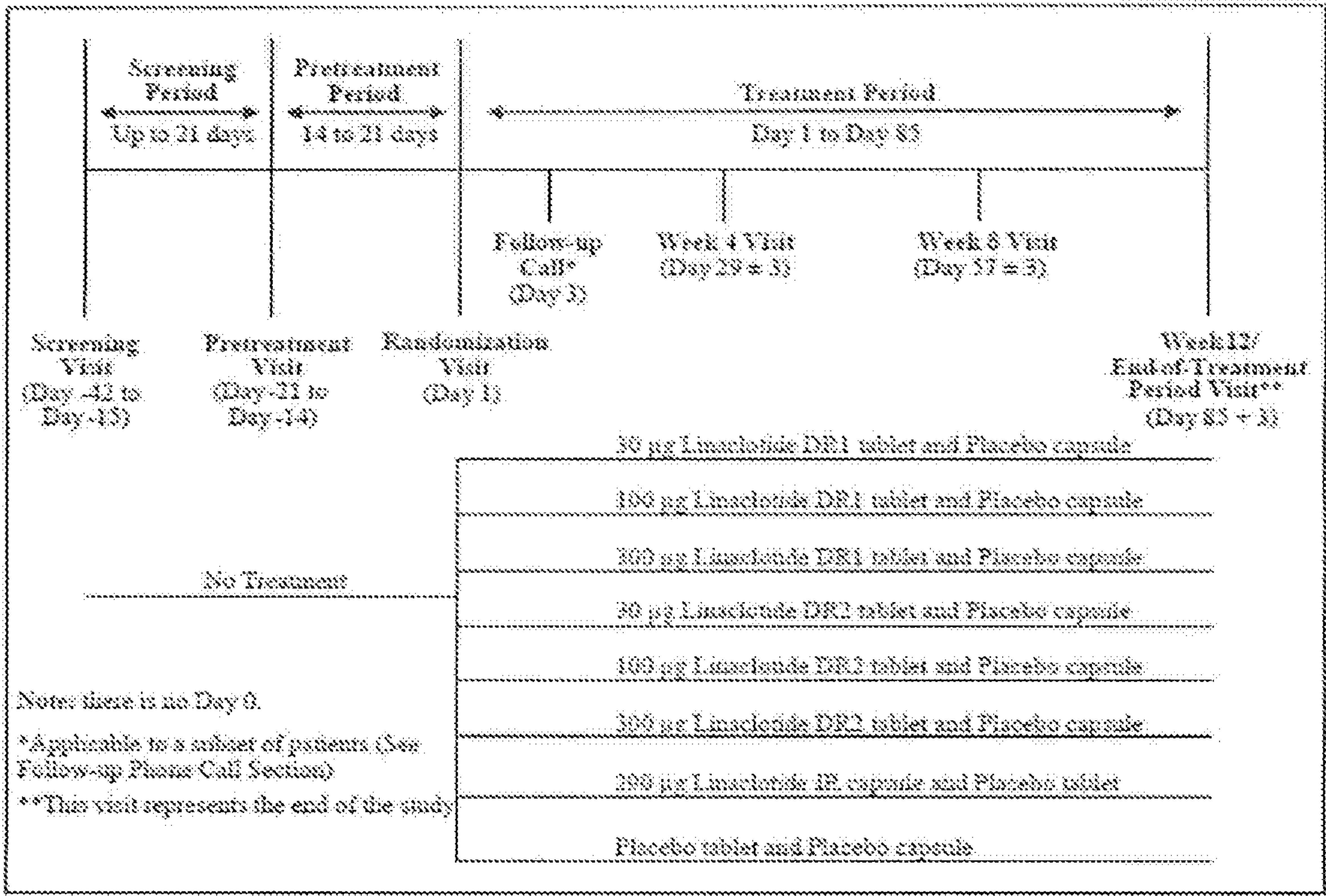
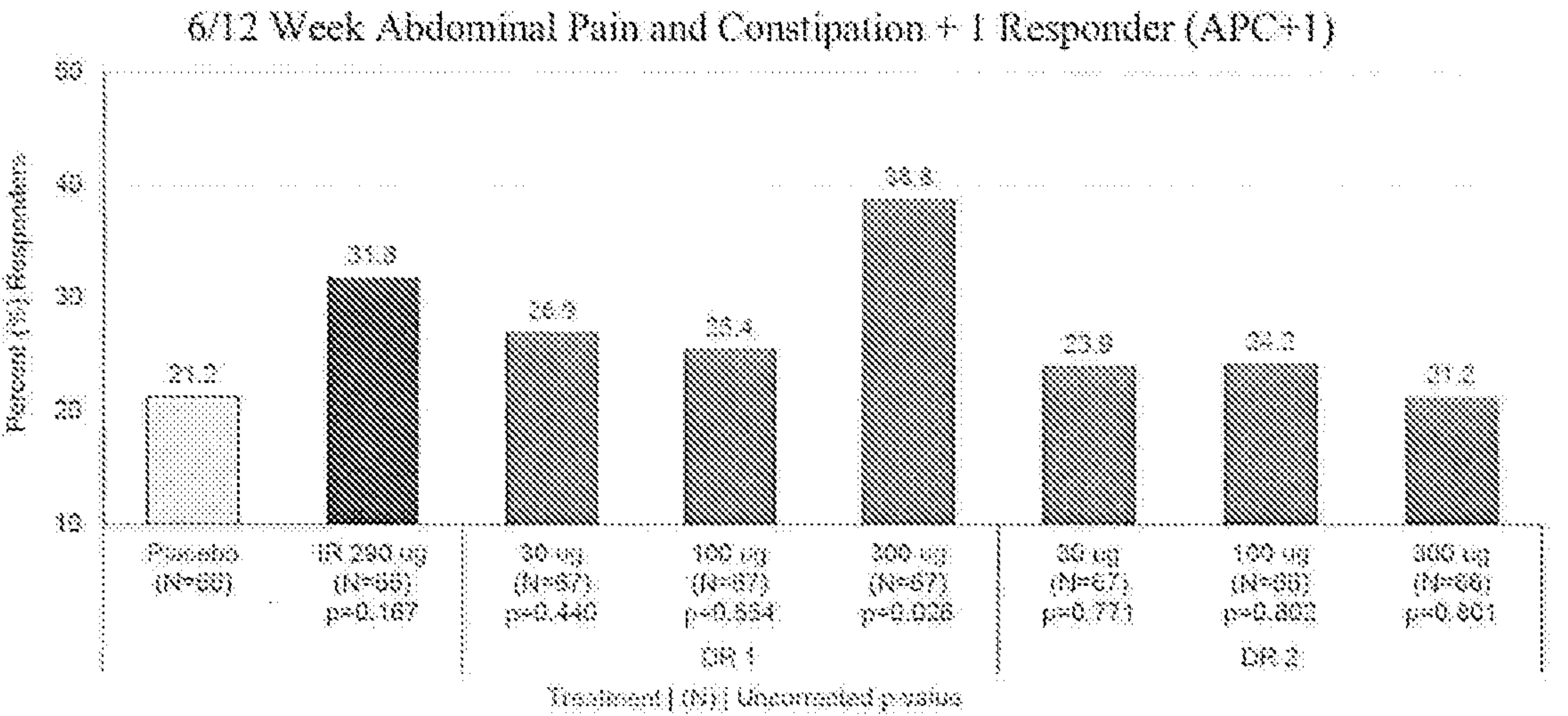


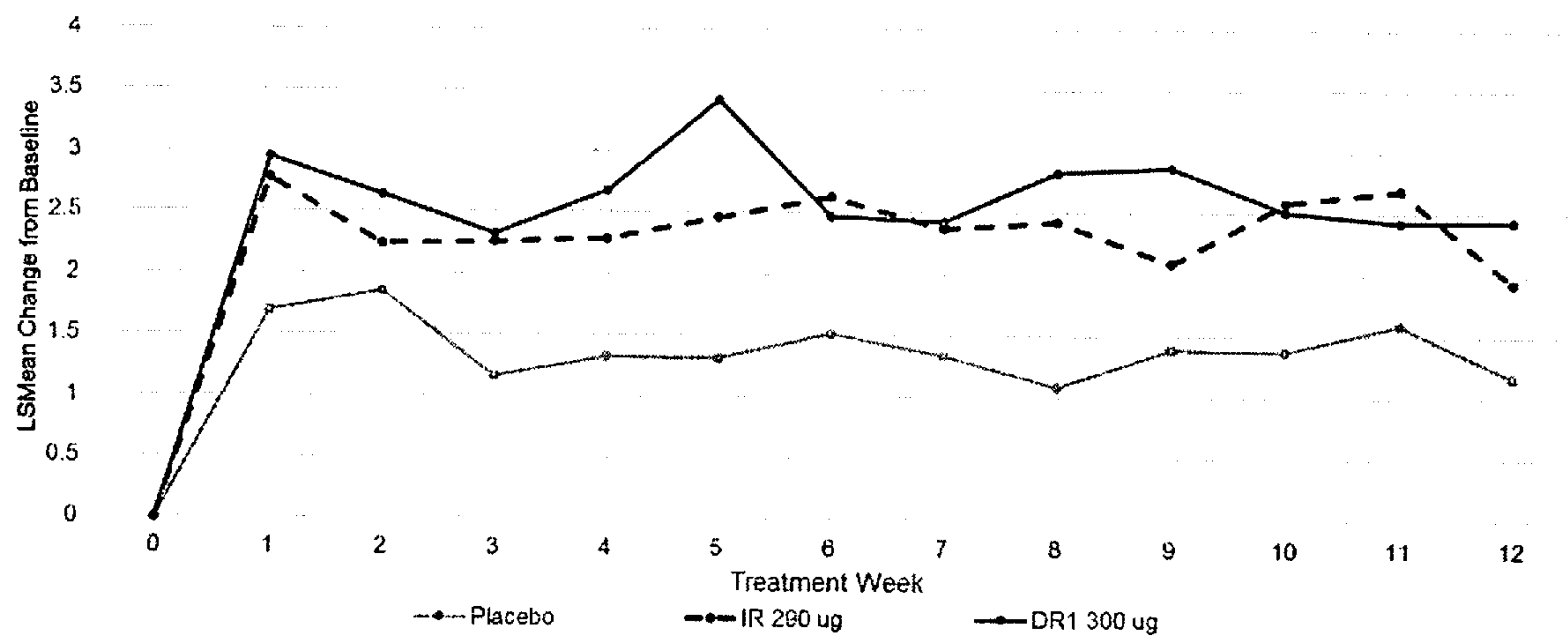
FIG. 1



P-values vs. placebo based on CMH test controlling for geographic region; not corrected for multiplicity.
DR1 Correlation Test p=0.025; DR2 Correlation Test p=0.858.

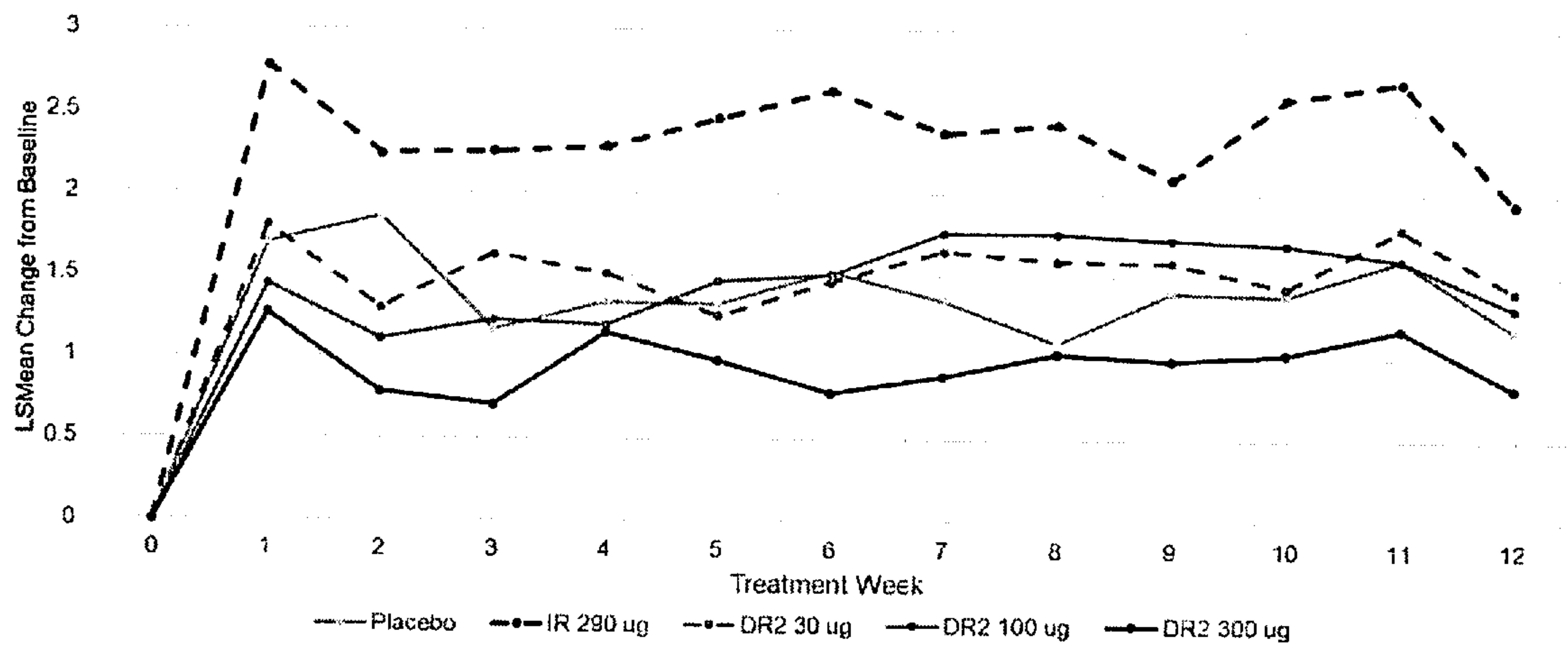
FIG. 2

SBM: Change from Baseline by Week: DR1



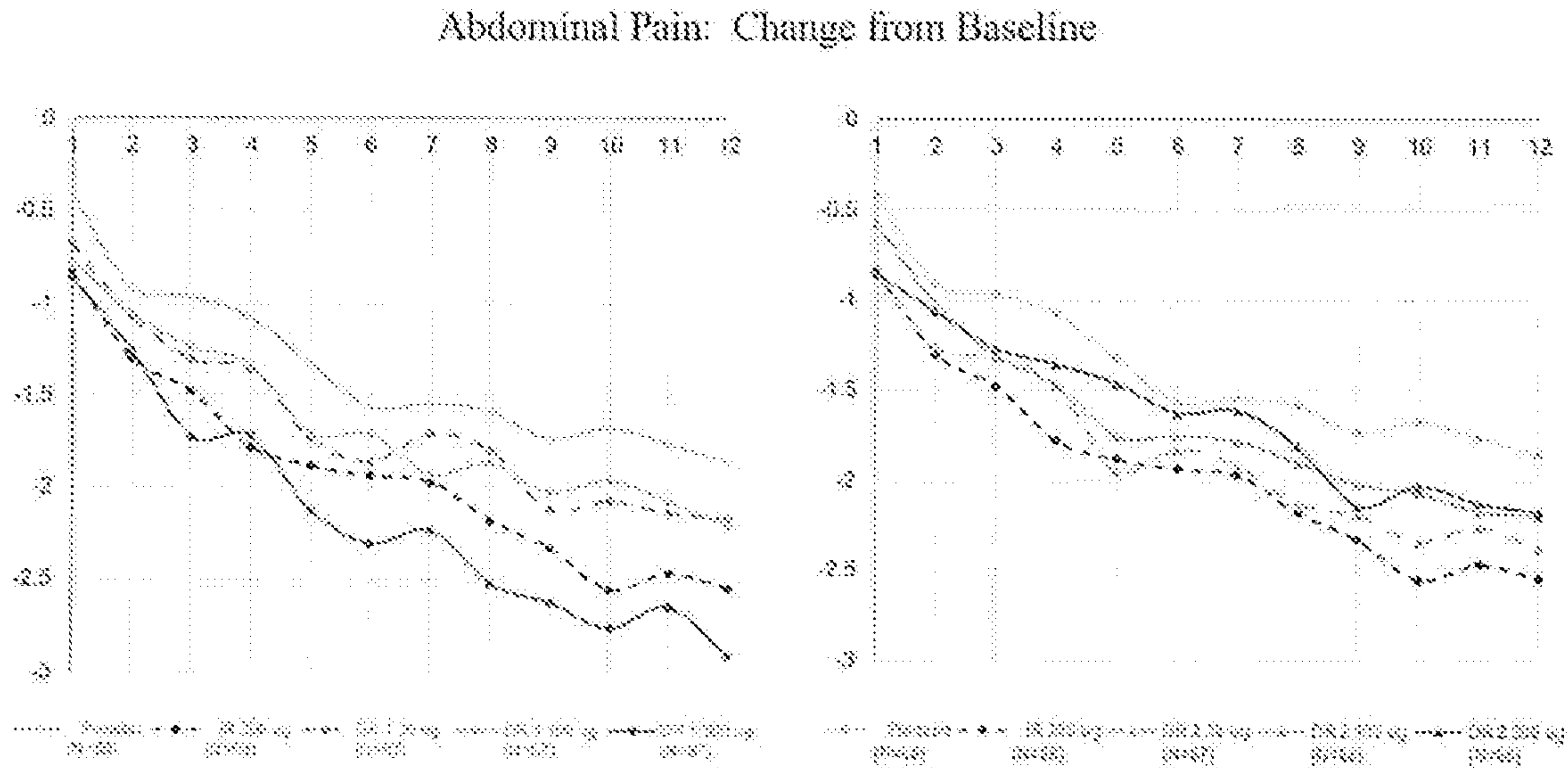
LSMeans based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate.

FIG. 3A



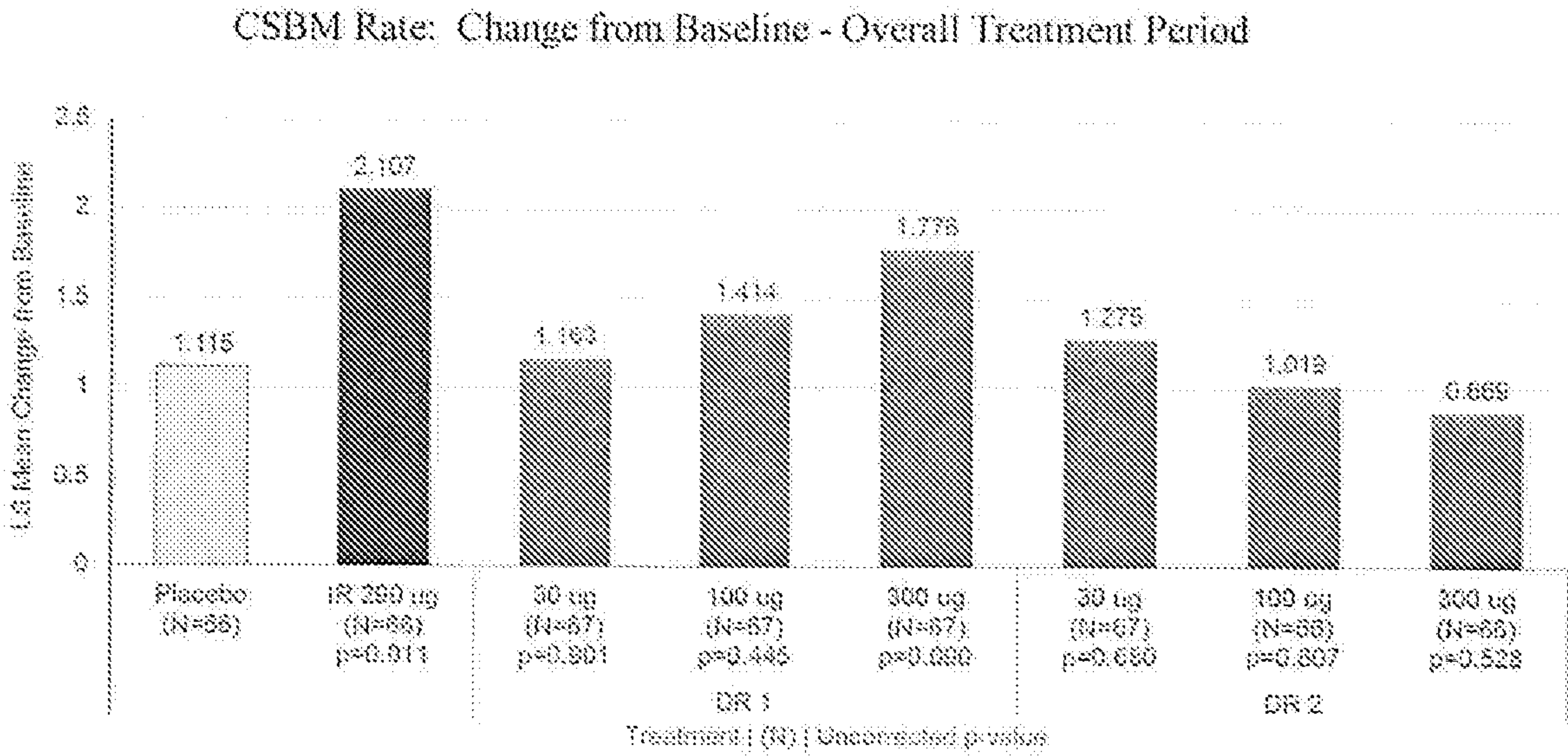
LSMeans based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate.

FIG. 3B



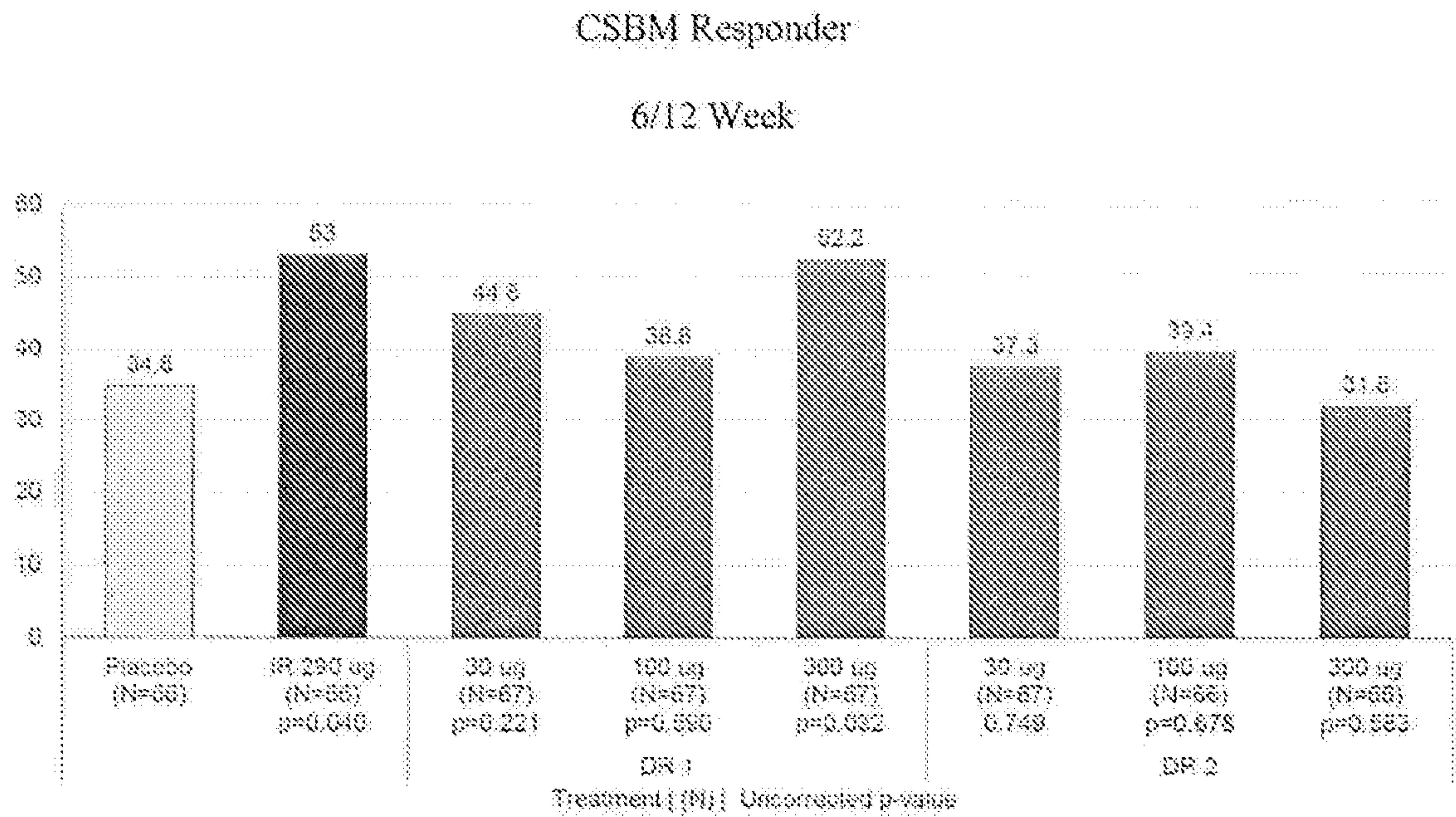
P-values vs. placebo and LS means based on overall treatment effect from MMRM with treatment, week, region, and treatment by week as fixed effects and baseline as a covariate. Compound symmetry error structure used to model intra-subject correlation. P-values not corrected for multiplicity.

FIG. 4



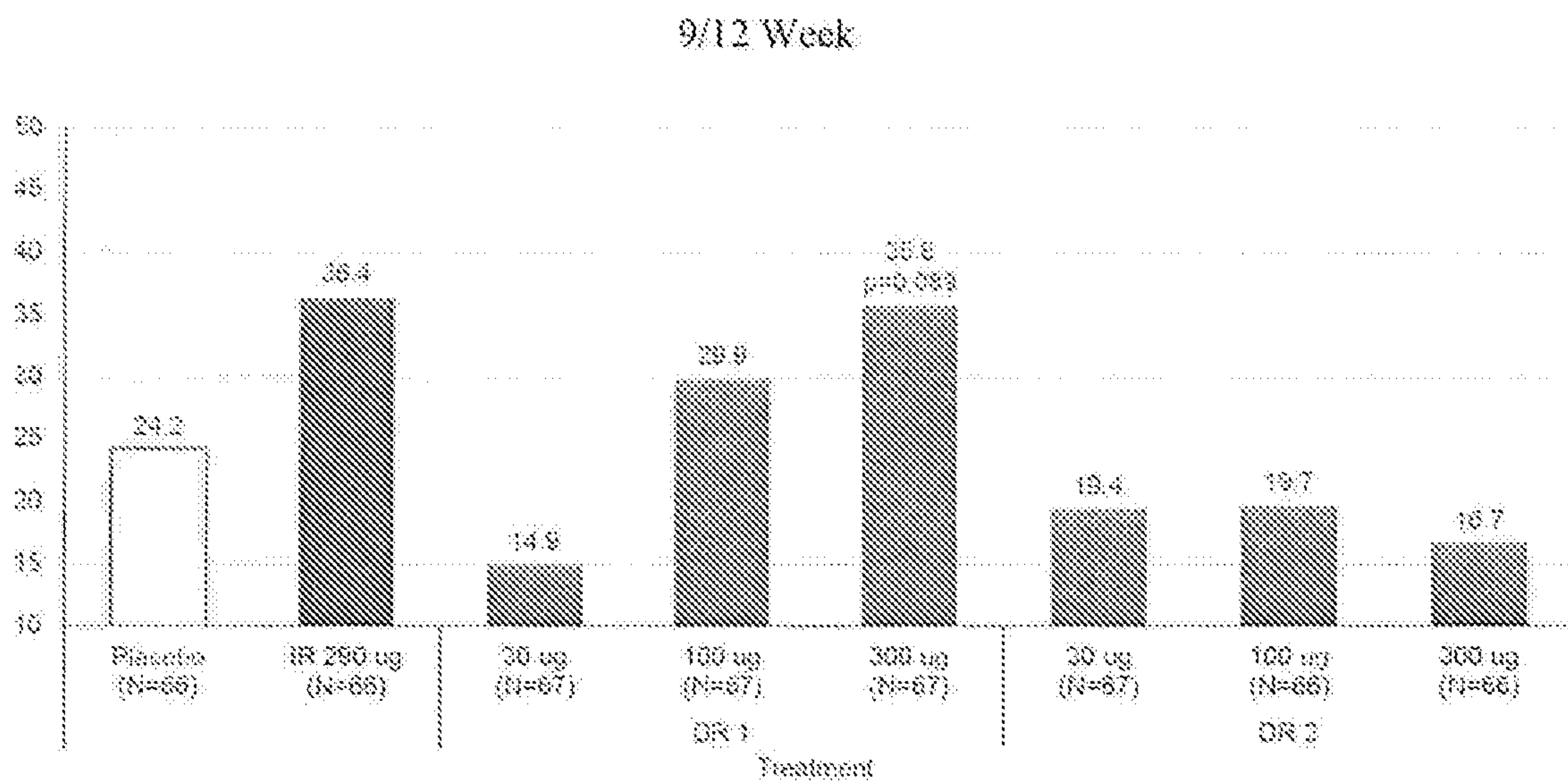
P-values vs. placebo and LS means based on overall treatment effect from MMRM with treatment, week, region, and treatment by week as fixed effects and baseline as a covariate. Compound symmetry error structure used to model intra-subject correlation. P-values not corrected for multiplicity.
DR1 Linear Trend Test: p=0.070; DR2 Linear Trend Test: p=0.420.

FIG. 5



P-values vs. Placebo based on CMH test controlling for geographic region; not corrected for multiplicity.

FIG. 6A



P-values vs. Placebo based on CMH test controlling for geographic region; not corrected for multiplicity or multiple comparisons.

FIG. 6B

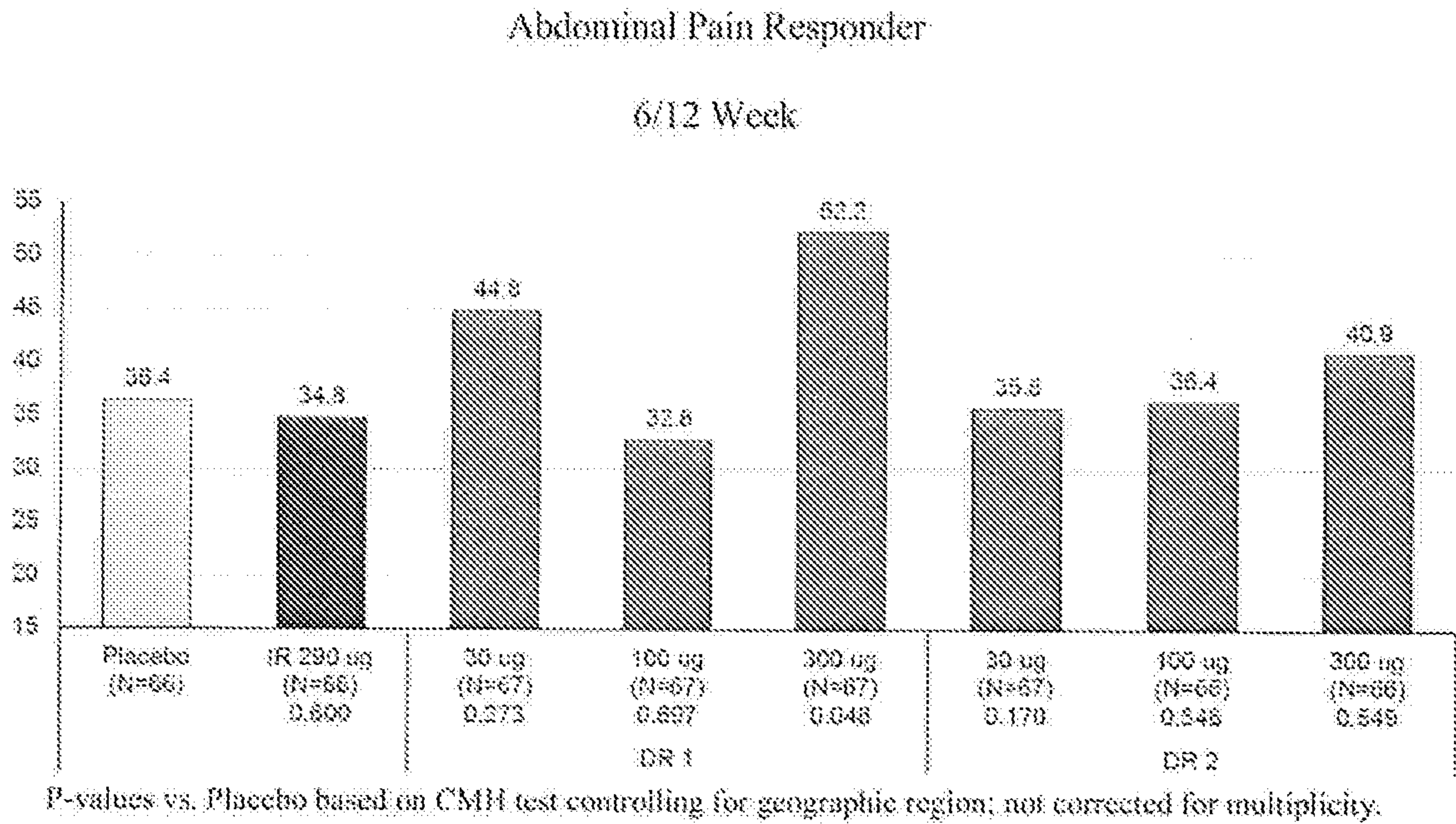


FIG. 7A

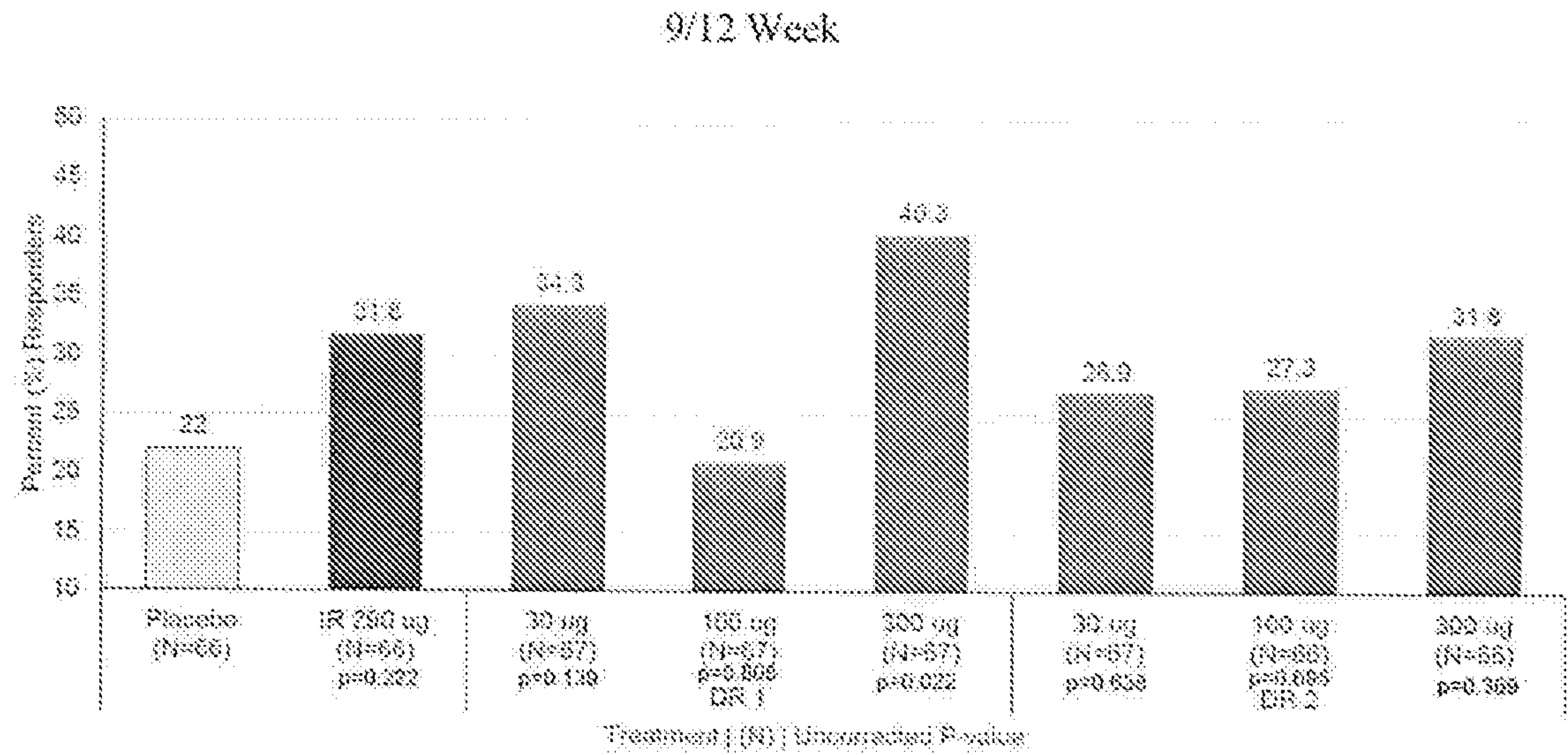
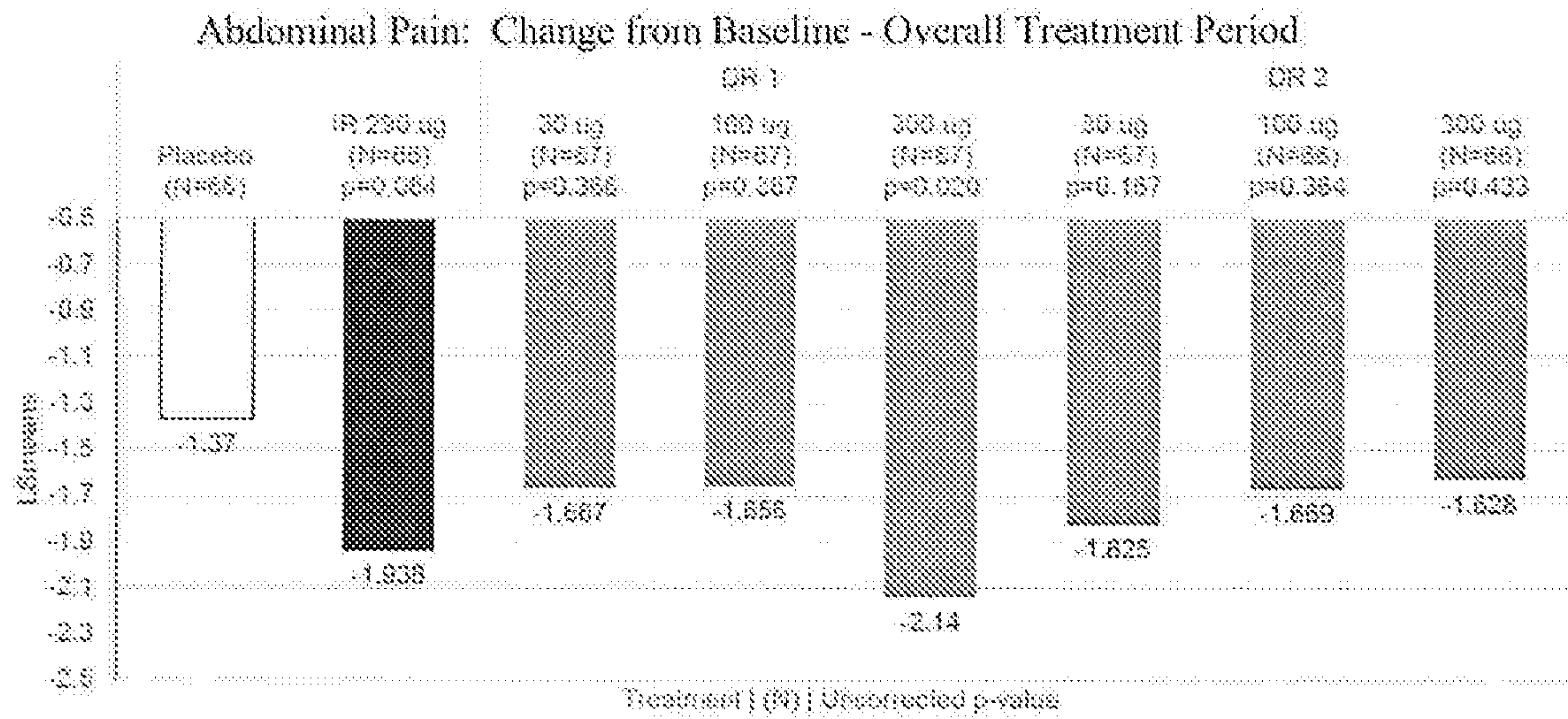


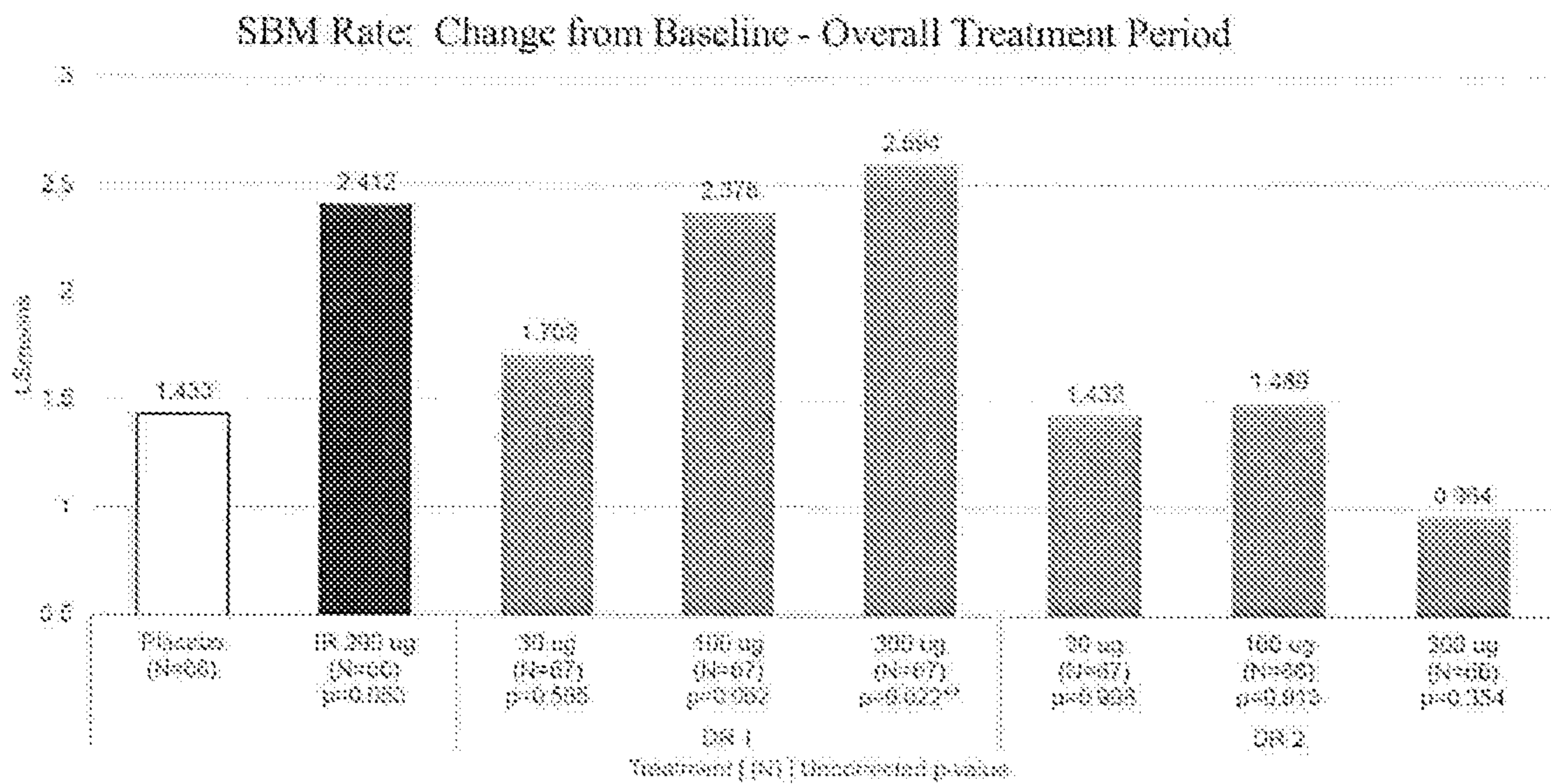
FIG. 7B



P-values vs. placebo and LS means based on overall treatment effect from MMRM with treatment, week, region, and treatment by week as fixed effects and baseline as a covariate. Compound symmetry error structure used to model intra-subject correlation. P-values not corrected for multiplicity.

DR1 Linear Trend Test: p=0.028; DR2 Linear Trend Test: p=0.553

FIG. 8

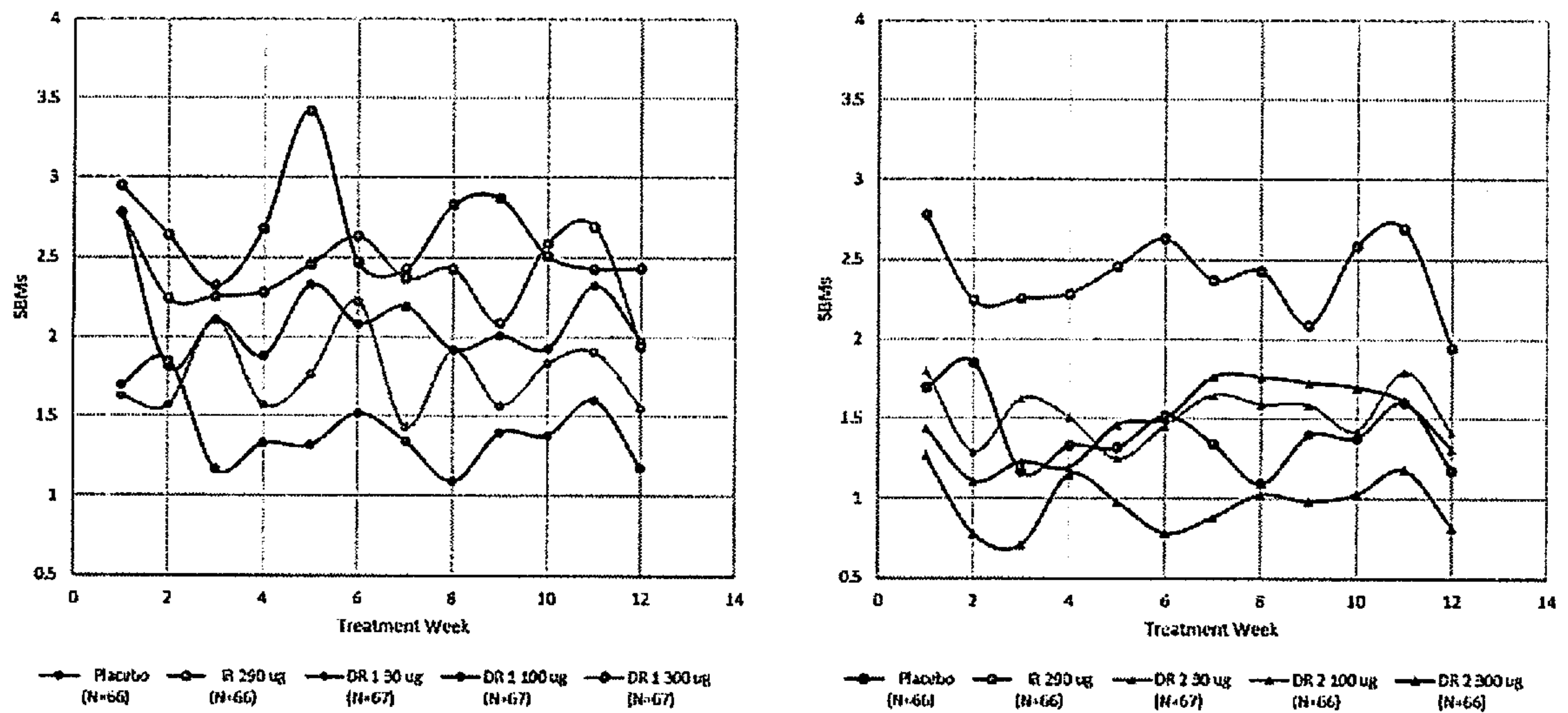


P-values vs. Placebo and LSMeans based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate.

P-values are not corrected for multiplicity.

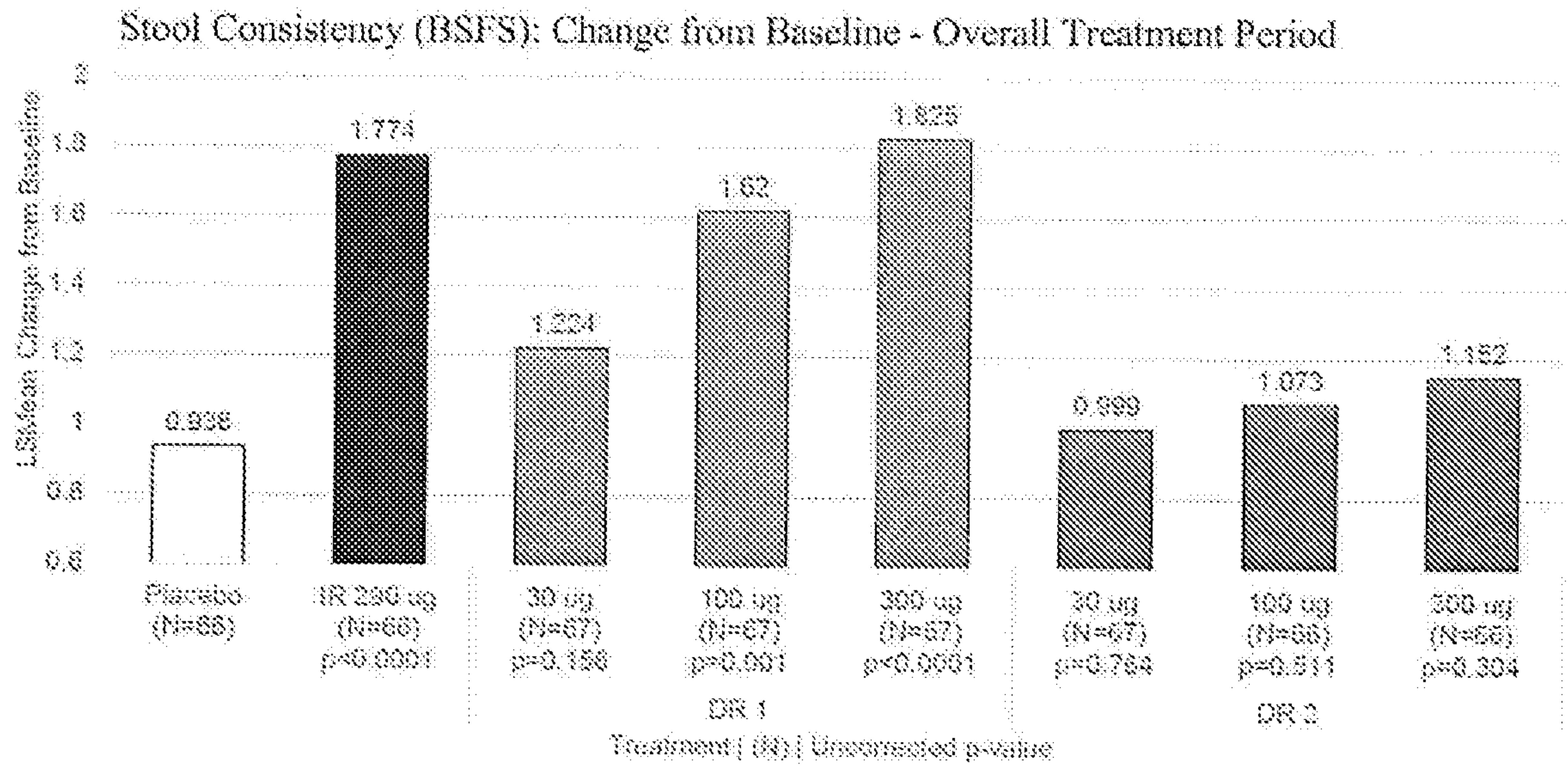
FIG. 9

SBM Rate: Change from Baseline



P-values vs. placebo and LSMeans based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate. P-values are not corrected for multiplicity nor multiple comparisons.
 $P < 0.10$: DR 1 30 ug – Week 3; DR 1 100 ug – Weeks 1, 3; DR 1 300 ug – Weeks 1, 3-5, 8-10-12; IR 290 ug – Weeks 1, 3, 5, 6, 8, 10.

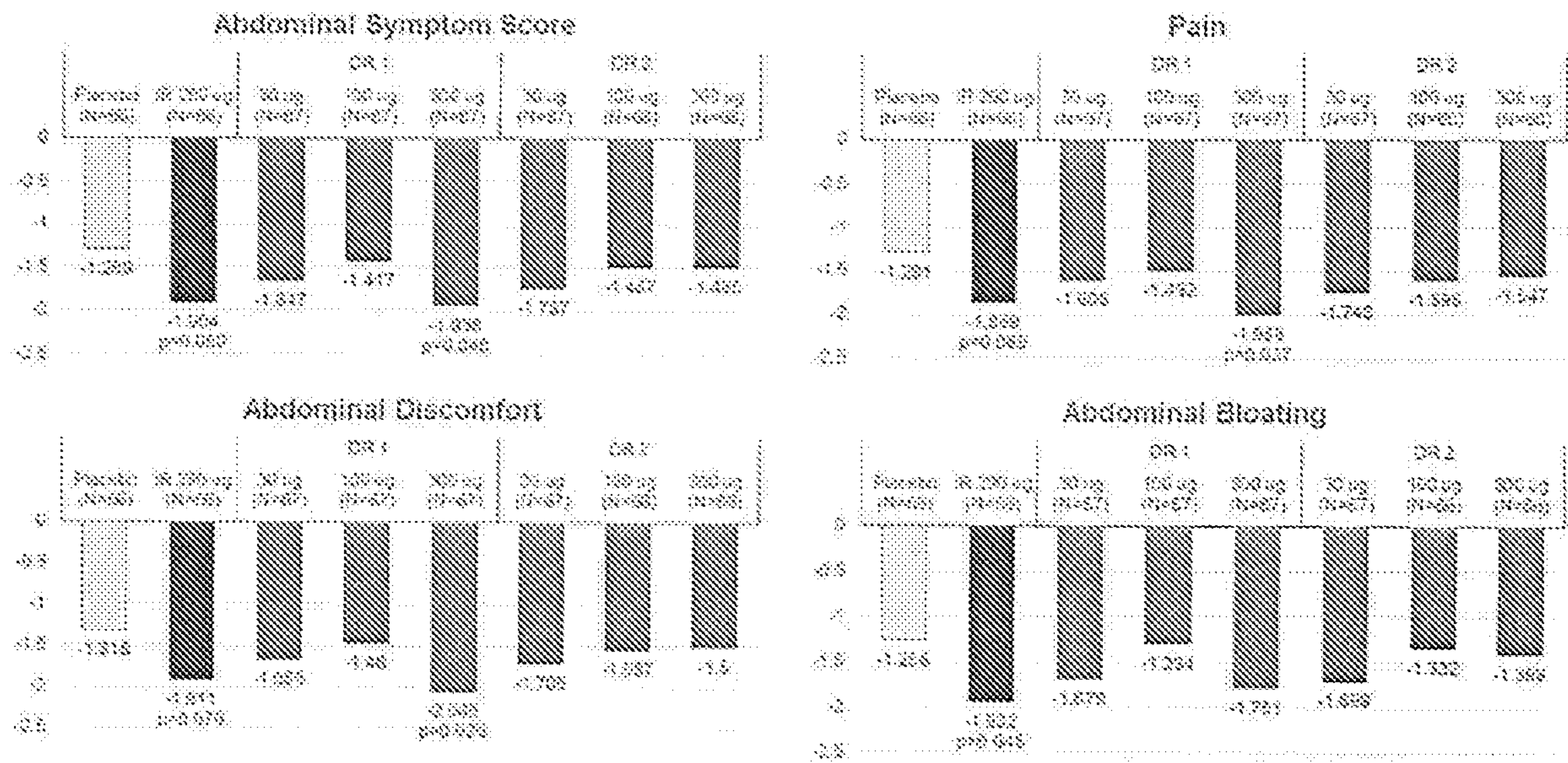
FIG. 10



P-values vs. placebo and LS means based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate. P-values are not corrected for multiplicity.

FIG. 11

Abdominal Symptom Score: Change from Baseline - Overall Treatment Period



P-values vs. Placebo and LS means based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate. P-values are not corrected for multiplicity.

FIG. 12

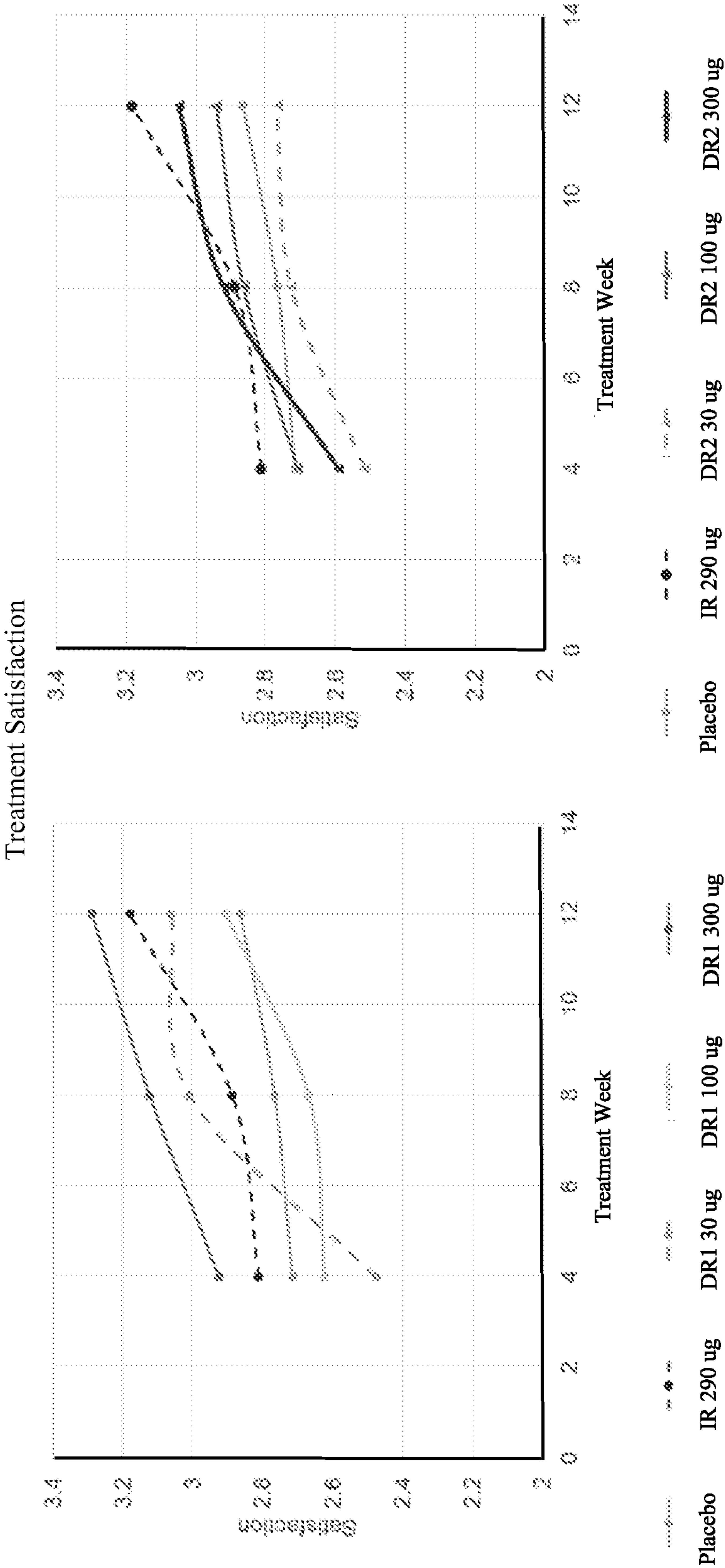


FIG. 13

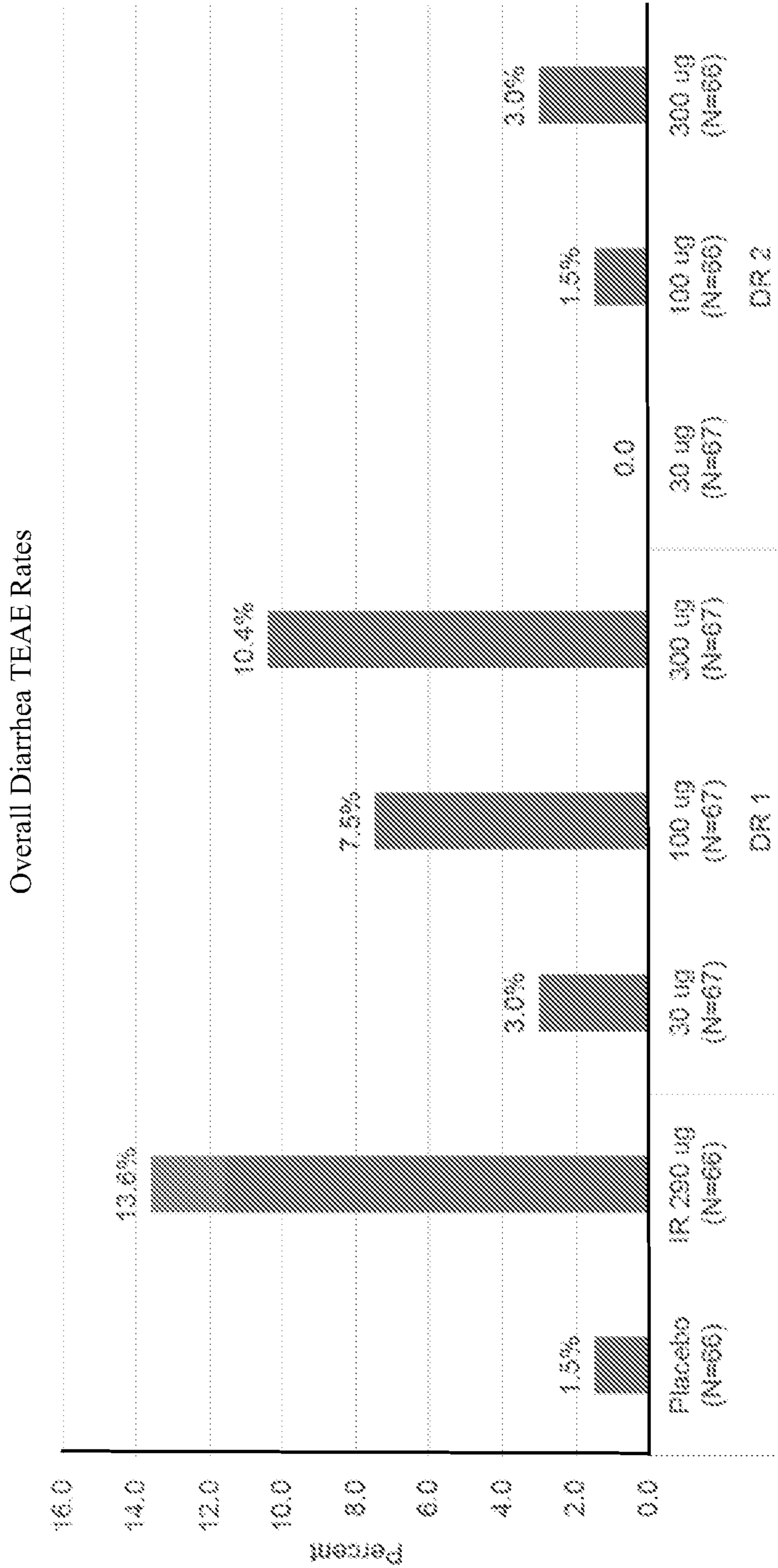


FIG. 14

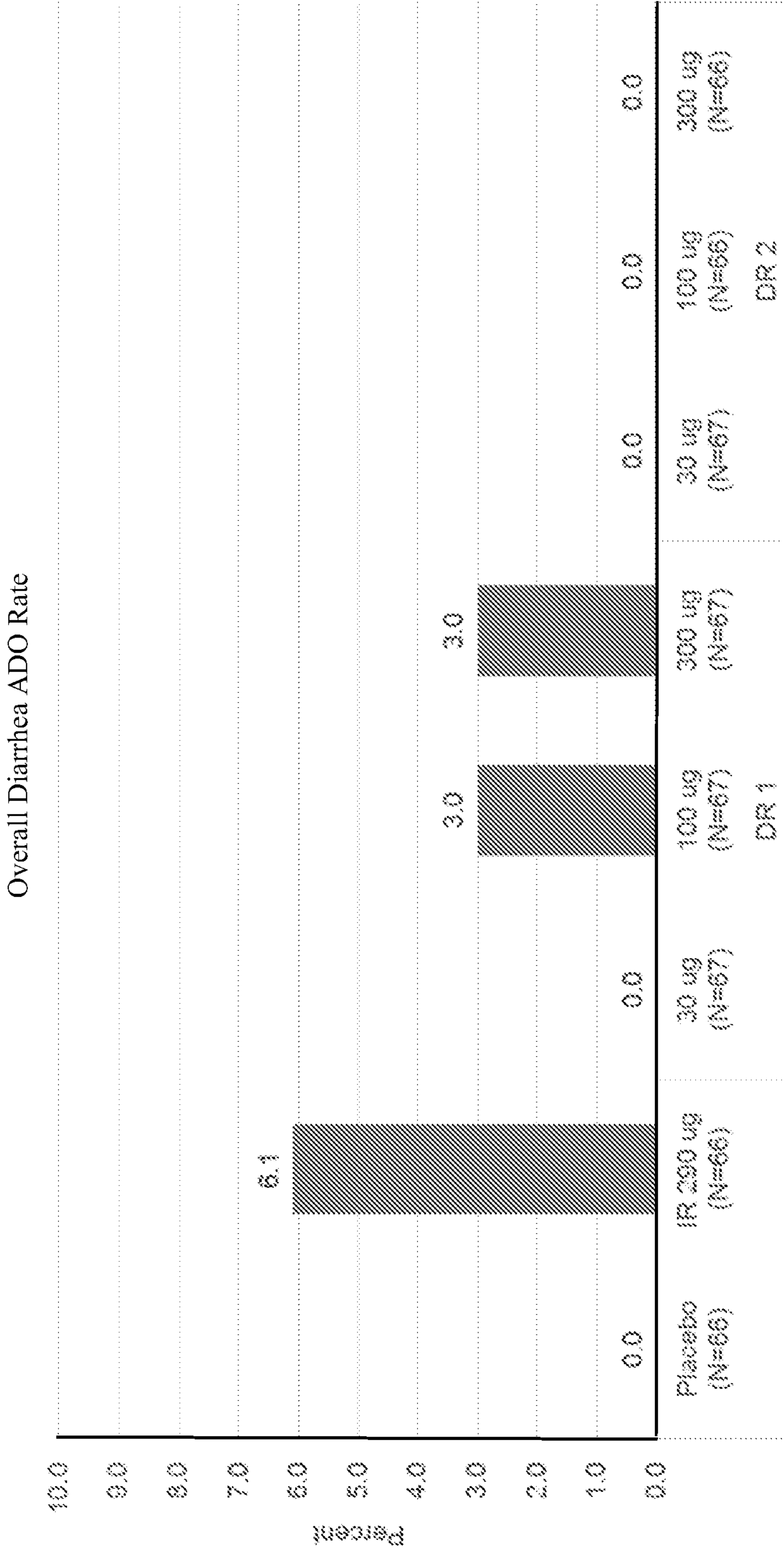


FIG. 15

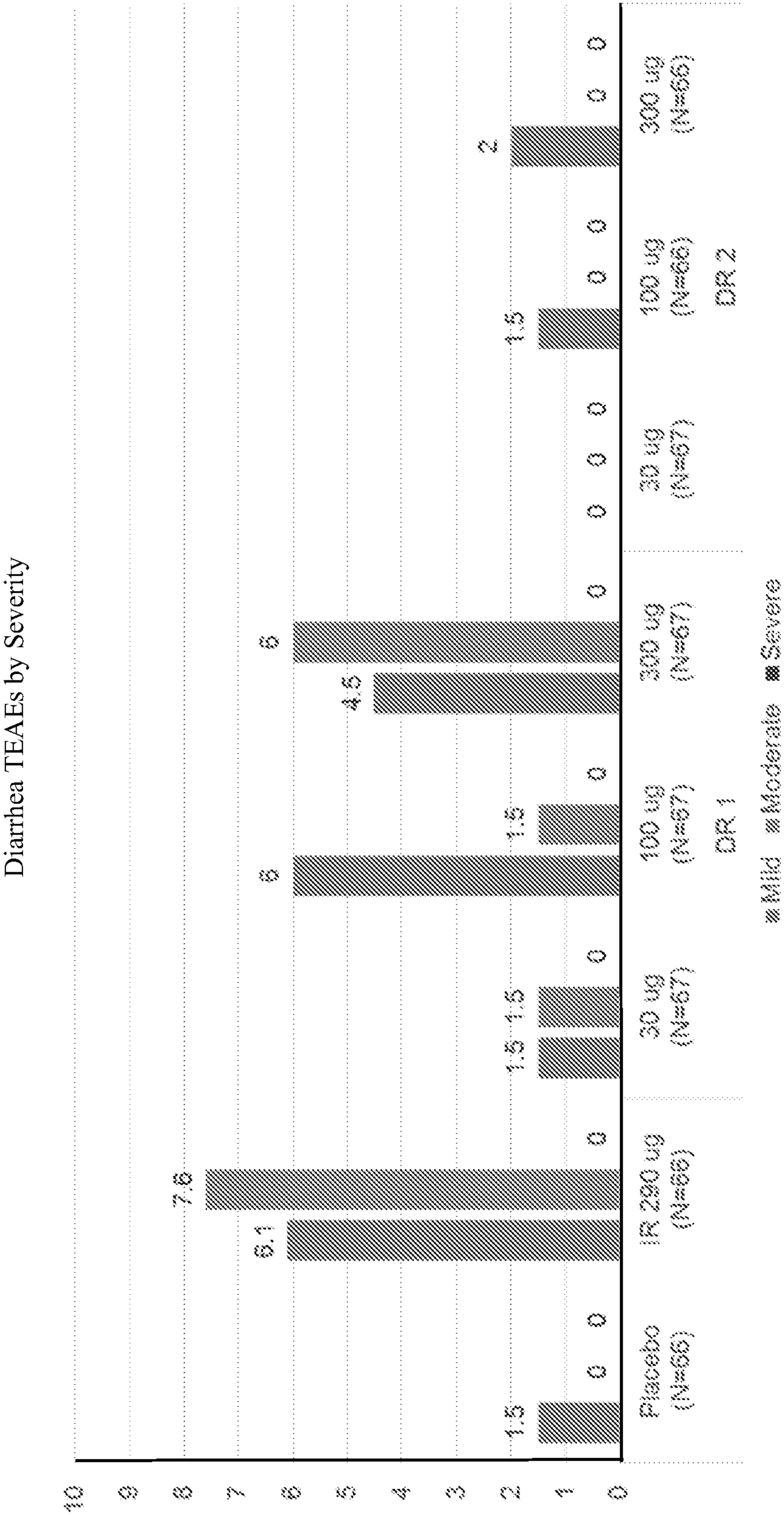


FIG. 16

Efficacy Parameters with Phase 3 Reference 6/12 Week Responder

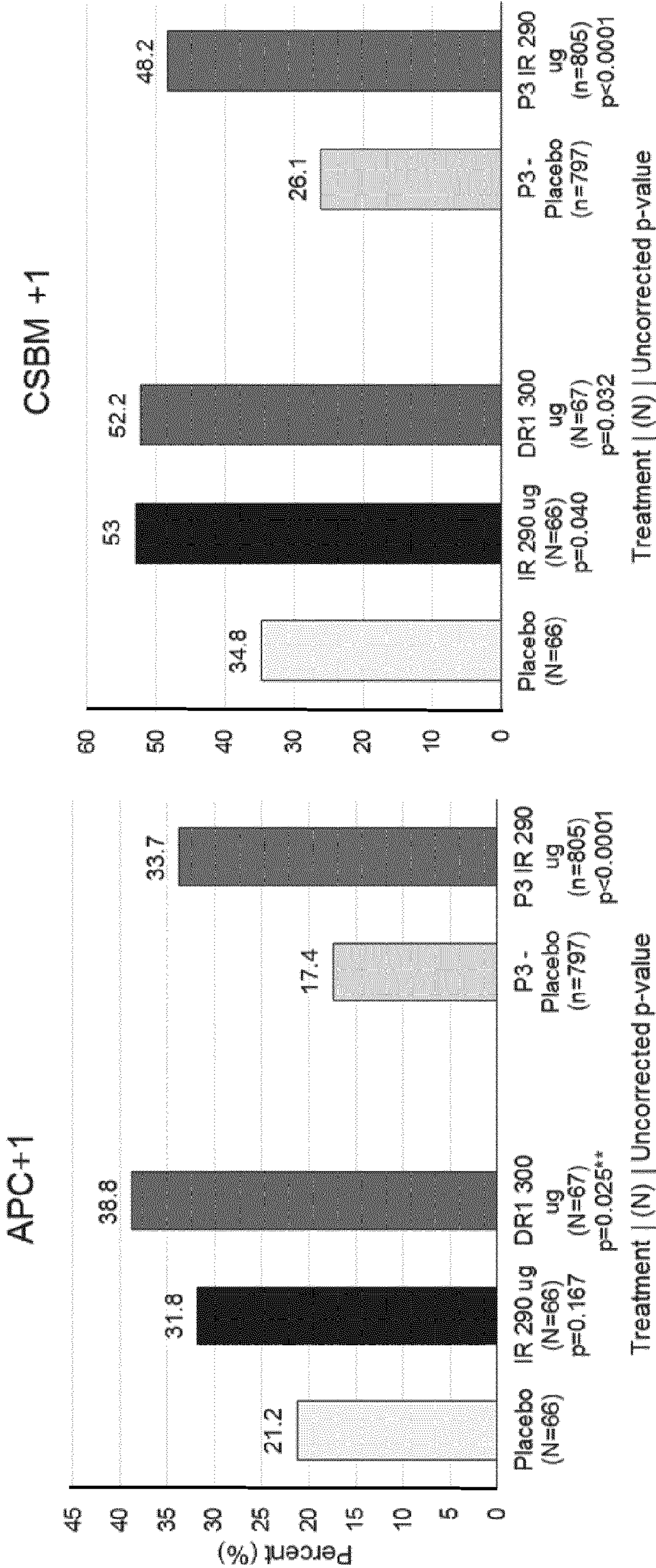


FIG. 17A

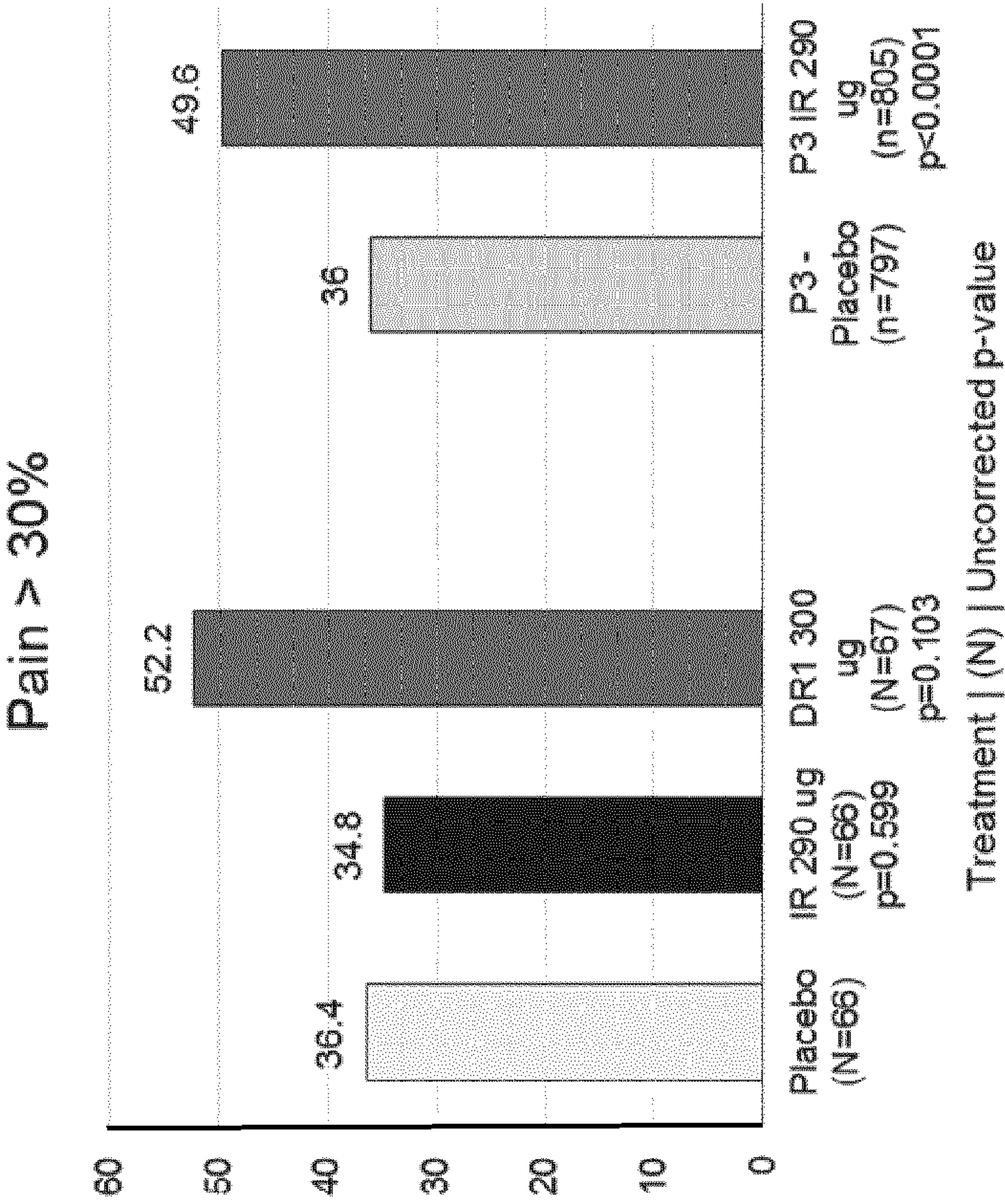


FIG. 17B

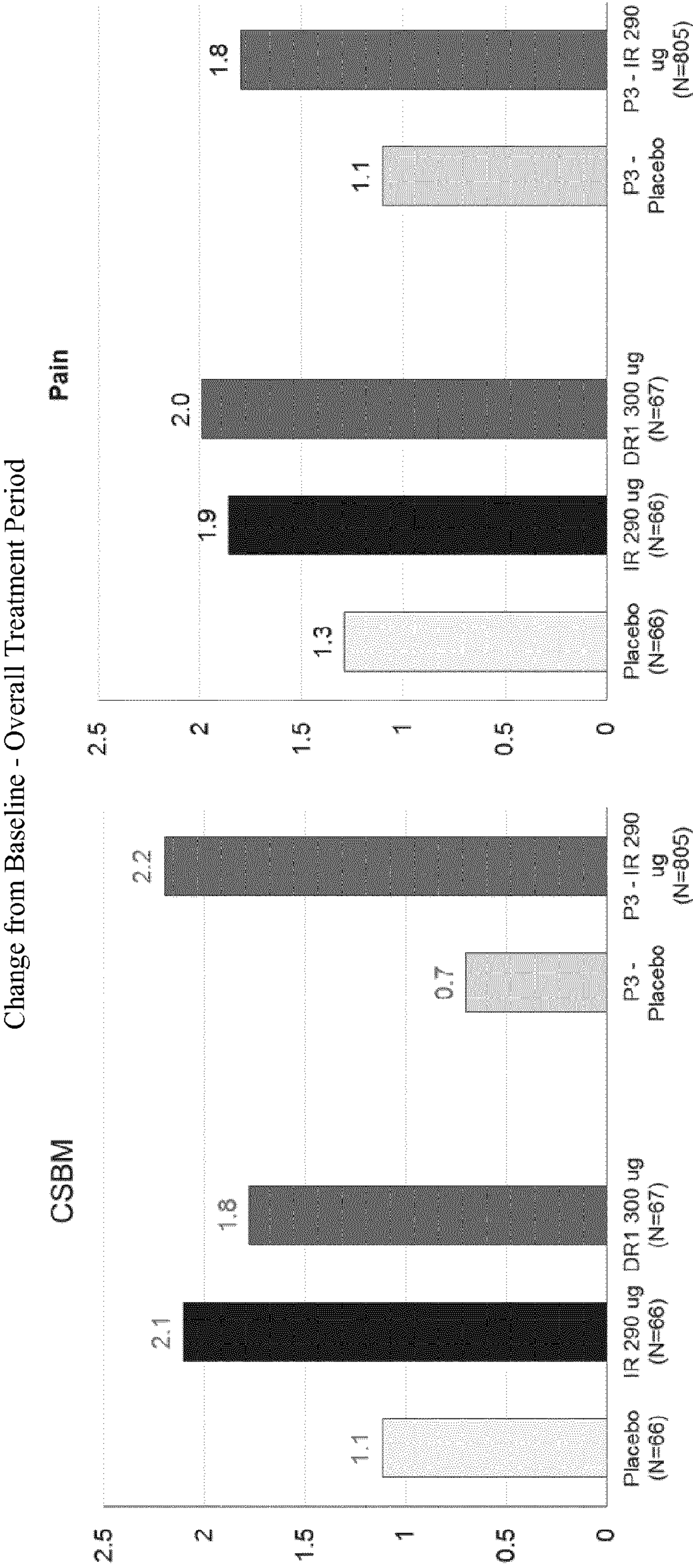


FIG. 17C

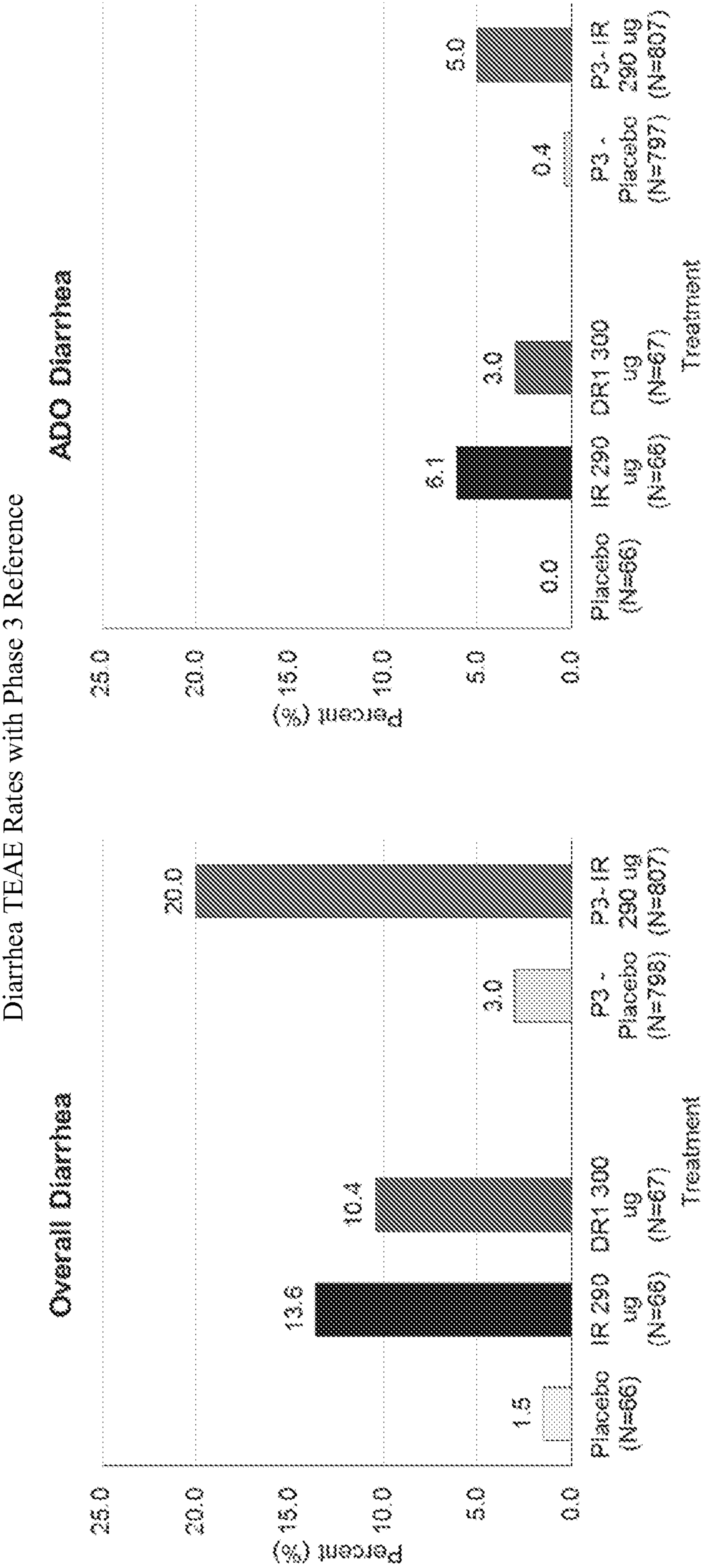


FIG. 18

Diarrhea TEAEs by Severity with Phase 3 Reference

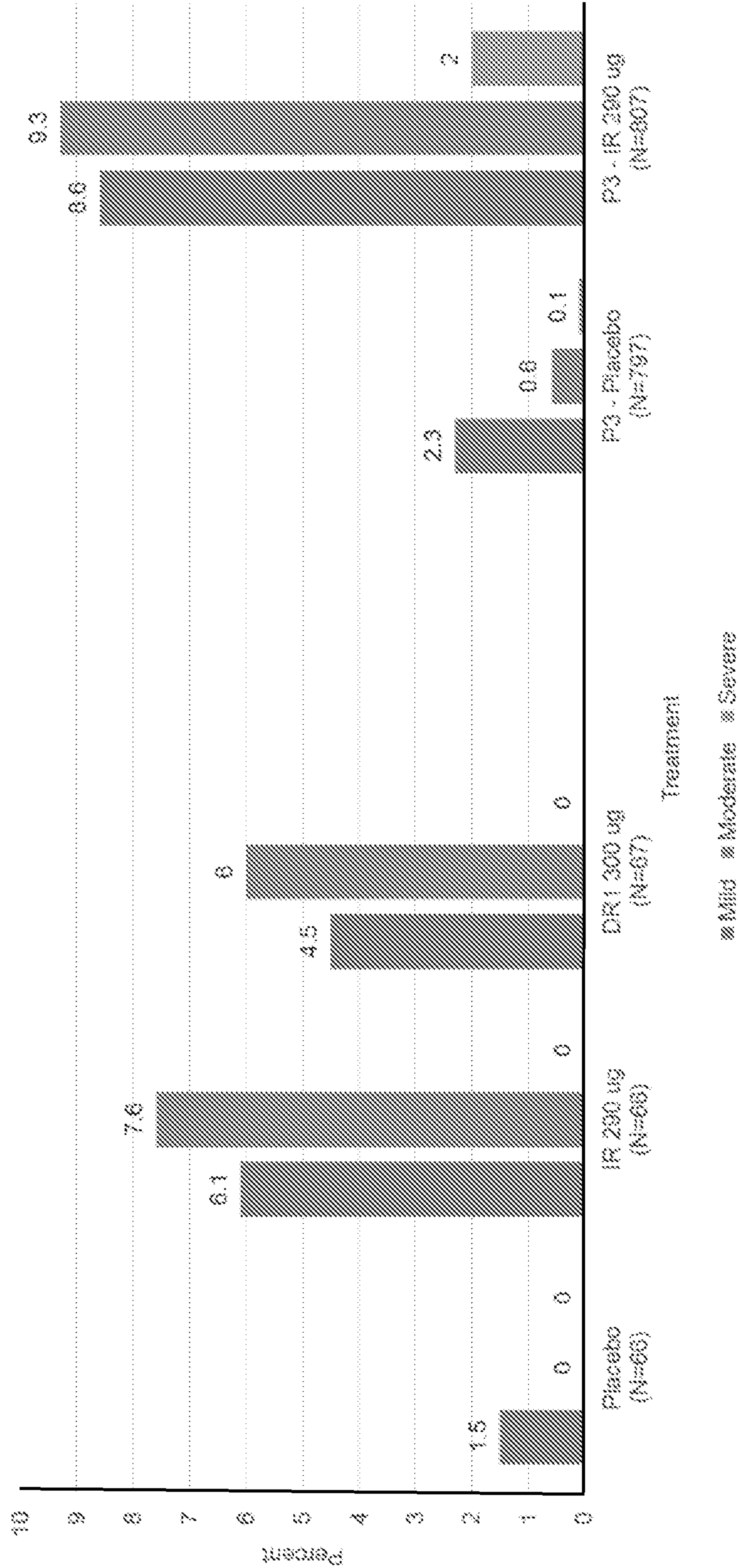


FIG. 19

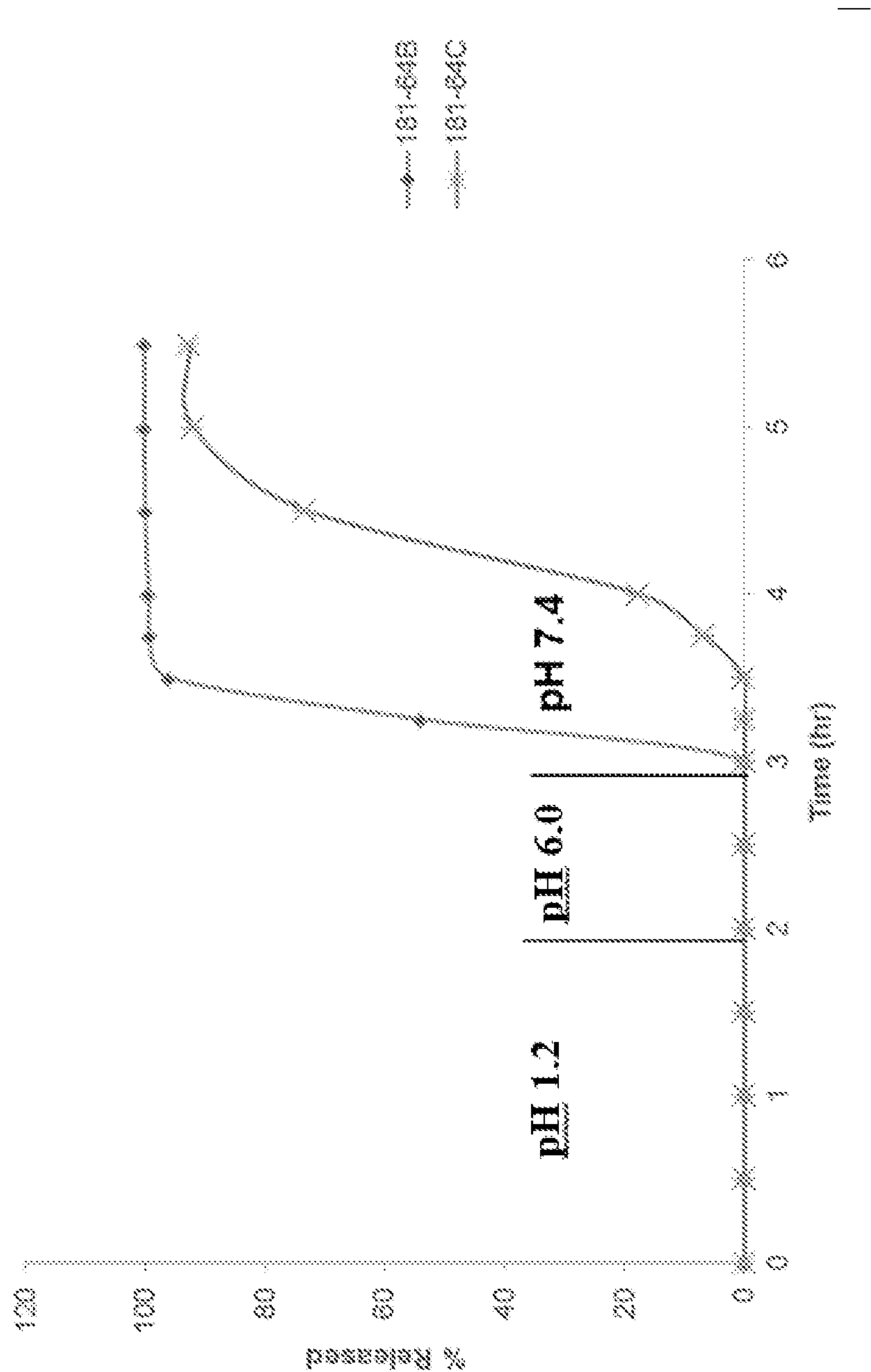


FIG. 20A

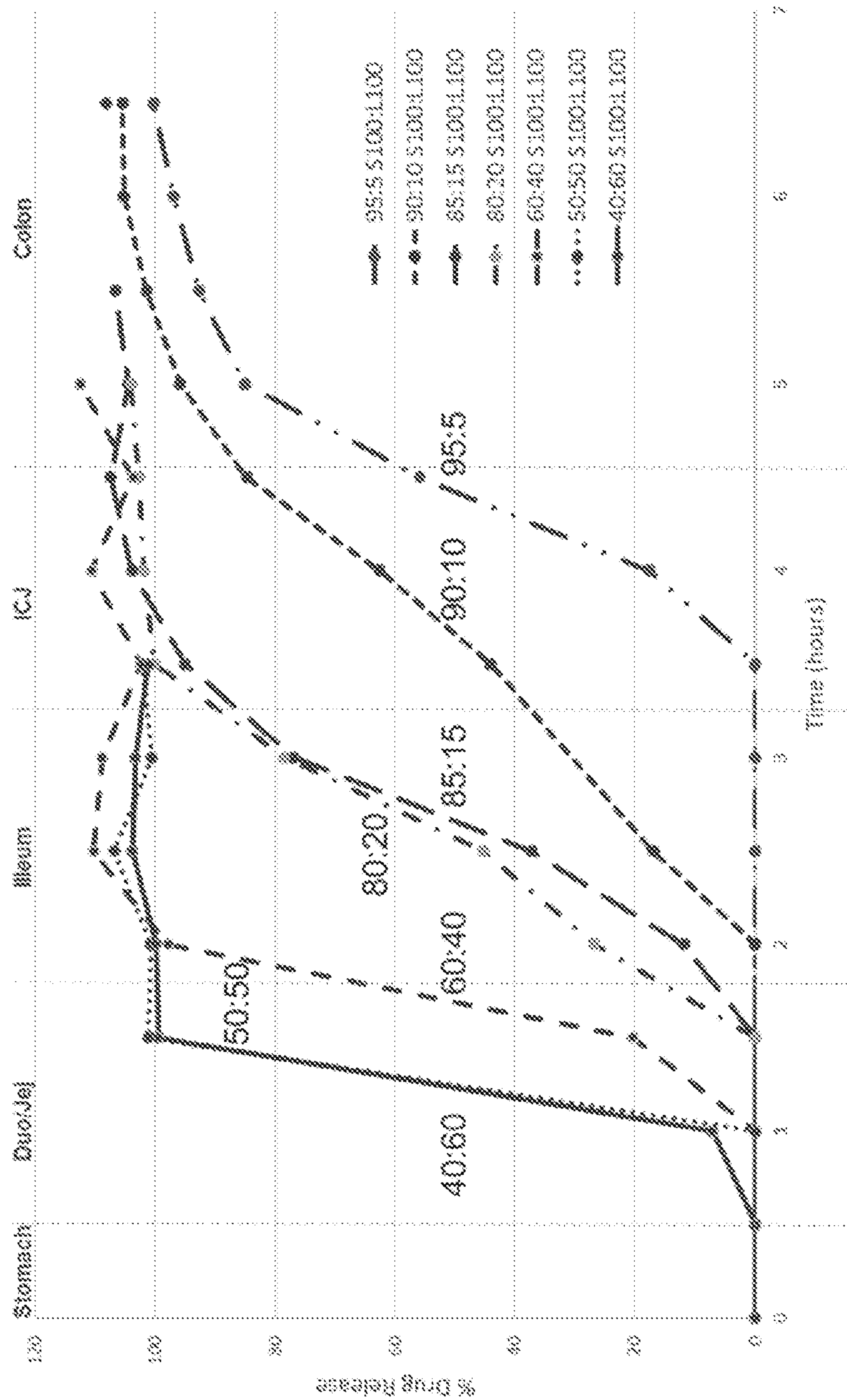
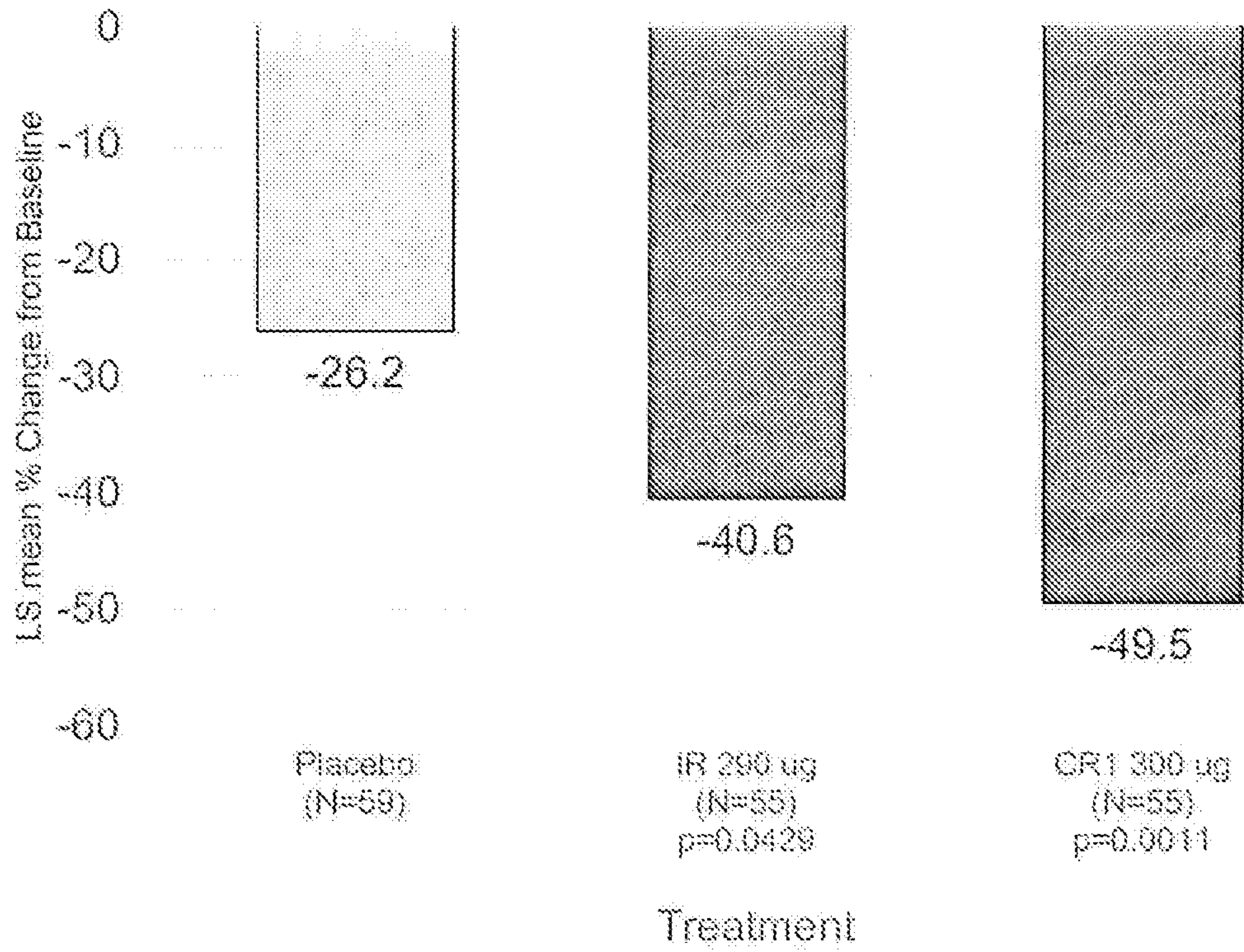


FIG. 20B



P-values vs. placebo and LSMeans based on an ANCOVA with treatment and region as fixed effect and baseline as a covariate. P-values are not corrected for multiplicity.

FIG. 21