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## (54) Title: METHODS OF TREATING IRRITABLE BOWEL SYNDROME

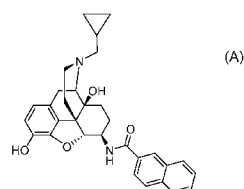
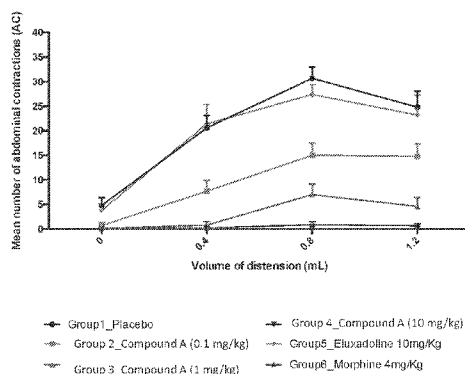


FIG. 3

Data on CRD following WRS sensitization



(57) Abstract: The disclosure provides methods for treating irritable bowel syndrome, including a symptom of irritable bowel syndrome with Compound A: or a pharmaceutically acceptable salt thereof.

**Declarations under Rule 4.17:**

- *as to applicant's entitlement to apply for and be granted a patent (Rule 4.17(ii))*

**Published:**

- *with international search report (Art. 21(3))*

## METHODS OF TREATING IRRITABLE BOWEL SYNDROME

### RELATED APPLICATIONS

[001] The application claims priority to, and the benefit of, U.S. Provisional Application No. 63/136,869, filed on January 13, 2021, the contents of which are incorporated herein by reference in their entirety.

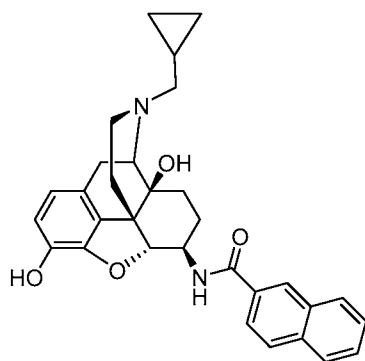
### BACKGROUND

[002] Irritable bowel syndrome (IBS) is a functional intestinal disorder characterized by abnormal bowel habits and chronic abdominal pain due to visceral hypersensitivity in the absence of inflammation. Colonic hypersensitivity without overt inflammation of the colon can be induced by intracolonic infusion of dilute acetic acid (AA). Compound A is an analgesic that activates a CNS target (e.g., the mu-kappa heteromer) and may be capable of reducing colonic hypersensitivity and serve as a treatment for subjects with IBS.

[003] The disclosure arises from a need to provide further compounds for the treatment of irritable bowel syndrome (IBS). In particular, compounds with improved physicochemical, pharmacological and pharmaceutical properties to existing compounds are desirable.

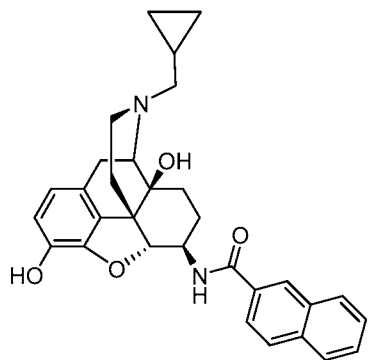
### SUMMARY

[004] In some aspects, the present disclosure provides a method of treating or preventing irritable bowel syndrome in a subject, comprising administering to the subject a therapeutically effective

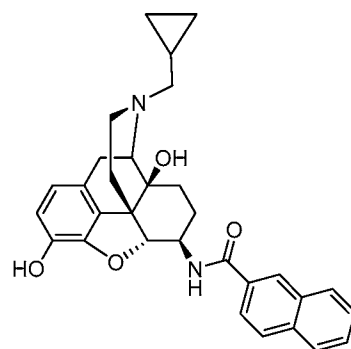


amount of Compound A, (A), or a pharmaceutically acceptable salt thereof.

[005] In some aspects, the present disclosure provides a method of treating or preventing irritable bowel syndrome in a subject, comprising administering to the subject Compound A,

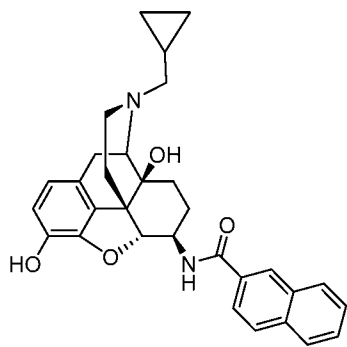


(A), or a pharmaceutically acceptable salt thereof.



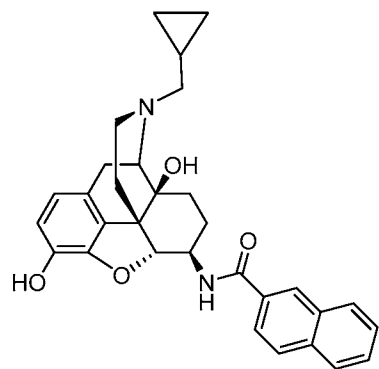
[006] In some aspects, the present disclosure provides Compound A, (A), or a pharmaceutically acceptable salt thereof, for use in treating or preventing irritable bowel syndrome.

[007] In some aspects, the present disclosure provides use of Compound A,



(A), or a pharmaceutically acceptable salt thereof, in the manufacture of a medicament for treating or preventing irritable bowel syndrome.

[008] In some aspects, the present disclosure provides use of Compound A,



(A), or a pharmaceutically acceptable salt thereof, for the treatment or prevention of irritable bowel syndrome.

[009] Unless otherwise defined, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. In the specification, the singular forms also include the plural unless the context clearly dictates otherwise. Although methods and materials similar or equivalent to those described herein can be used in the practice or testing of the present disclosure, suitable methods and materials are described below. All publications, patent applications, patents and other references mentioned herein are incorporated by reference. The references cited herein are not admitted to be prior art to the claimed invention. In the case of conflict, the present specification, including definitions, will control. In addition, the materials, methods and examples are illustrative only and are not intended to be limiting. In the case of conflict between the chemical structures and names of the compounds disclosed herein, the chemical structures will control.

[010] Other features and advantages of the disclosure will be apparent from the following detailed description and claims.

#### BRIEF DESCRIPTION OF THE DRAWINGS

[011] The patent or application file contains at least one drawing executed in color. Copies of this patent or patent application publication with color drawings will be provided by the Office upon request and payment of the necessary fee.

[012] **FIG. 1A** and **FIG. 1B** depict the effect of locally administered Compound A on acetic acid-induced colonic hypersensitivity, wherein A.A. denotes Acetic Acid.

[013] **FIG. 2** depicts the wrap restraint stress model.

[014] **FIG. 3** depicts the complete reduction data in abdominal contractions for the wrap restrain stress model with placebo, Compound A at 0.1 mg/kg, 1 mg/kg and 10 mg/kg administered intracolonic, morphine at 4 mg/kg SC, and Eluxadoline at 10 mg/kg, PO.

[015] **FIG. 4** depicts immunofluorescence staining of mu-kappa opioid receptor heteromers in rat colon tissue.

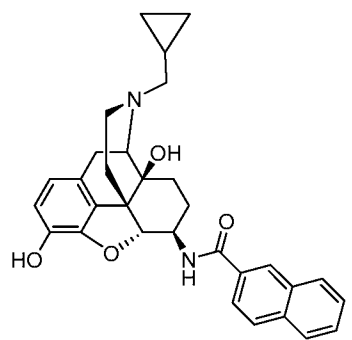
#### DETAILED DESCRIPTION

[016] The present disclosure relates to the use of Compound A, or a pharmaceutically acceptable salt thereof, in the treatment of irritable bowel syndrome.

#### *Definitions*

[017] Unless otherwise stated, the following terms used in the specification and claims have the following meanings set out below.

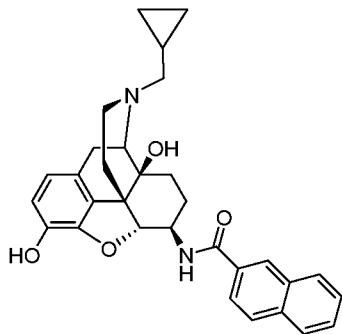
[018] Unless explicitly indicated otherwise, the terms “Compound A” and “N-((4aS,7R,7aR,12bS)-3-(cyclopropylmethyl)-4a,9-dihydroxy-2,3,4,4a,5,6,7,7a-octahydro-1H-4,12-methanobenzofuro[3,2-e]isoquinolin-7-yl)-2-naphthamide” refer to the compound which has the following structure:



[019] Compound A, and its preparation are disclosed in PCT Application Publication Nos. WO WO2010/006299. These publications are incorporated by reference herein in their entireties.

[020] Unless explicitly indicated otherwise, any reference to “Compound A”, and “N-((4aS,7R,7aR,12bS)-3-(cyclopropylmethyl)-4a,9-dihydroxy-2,3,4,4a,5,6,7,7a-octahydro-1H-4,12-methanobenzofuro[3,2-e]isoquinolin-7-yl)-2-naphthamide” refers also to pharmaceutically

acceptable salts of, and pharmaceutical compositions comprising, the compound which has the



following structure:

[021] As used herein, the expressions “one or more of A, B, or C,” “one or more A, B, or C,” “one or more of A, B, and C,” “one or more A, B, and C,” “selected from the group consisting of A, B, and C,” “selected from A, B, and C”, and the like are used interchangeably and all refer to a selection from a group consisting of A, B, and/or C, i.e., one or more As, one or more Bs, one or more Cs, or any combination thereof, unless indicated otherwise.

[022] It is to be understood that the present disclosure provides methods for the synthesis of the compounds of any of the Formulae described herein. The present disclosure also provides detailed methods for the synthesis of various disclosed compounds of the present disclosure according to the following scheme as well as those shown in the Examples.

[023] It is to be understood that, throughout the description, where compositions are described as having, including, or comprising specific components, it is contemplated that compositions also consist essentially of, or consist of, the recited components. Similarly, where methods or processes are described as having, including, or comprising specific process steps, the processes also consist essentially of, or consist of, the recited processing steps. Further, it should be understood that the order of steps or order for performing certain actions is immaterial so long as the invention remains operable. Moreover, two or more steps or actions can be conducted simultaneously.

[024] It is to be understood that the synthetic processes of the disclosure can tolerate a wide variety of functional groups, therefore various substituted starting materials can be used. The processes generally provide the desired final compound at or near the end of the overall process, although it may be desirable in certain instances to further convert the compound to a pharmaceutically acceptable salt thereof.

[025] It is to be understood that compounds of the present disclosure can be prepared in a variety of ways using commercially available starting materials, compounds known in the literature, or from readily prepared intermediates, by employing standard synthetic methods and procedures

either known to those skilled in the art, or which will be apparent to the skilled artisan in light of the teachings herein. Standard synthetic methods and procedures for the preparation of organic molecules and functional group transformations and manipulations can be obtained from the relevant scientific literature or from standard textbooks in the field. Although not limited to any one or several sources, classic texts such as Smith, M. B., March, J., *March's Advanced Organic Chemistry: Reactions, Mechanisms, and Structure*, 5<sup>th</sup> edition, John Wiley & Sons: New York, 2001; Greene, T.W., Wuts, P.G. M., *Protective Groups in Organic Synthesis*, 3<sup>rd</sup> edition, John Wiley & Sons: New York, 1999; R. Larock, *Comprehensive Organic Transformations*, VCH Publishers (1989); L. Fieser and M. Fieser, *Fieser and Fieser's Reagents for Organic Synthesis*, John Wiley and Sons (1994); and L. Paquette, ed., *Encyclopedia of Reagents for Organic Synthesis*, John Wiley and Sons (1995), incorporated by reference herein, are useful and recognized reference textbooks of organic synthesis known to those in the art

[026] One of ordinary skill in the art will note that, during the reaction sequences and synthetic scheme described herein, the order of certain steps may be changed, such as the introduction and removal of protecting groups. One of ordinary skill in the art will recognize that certain groups may require protection from the reaction conditions via the use of protecting groups. Protecting groups may also be used to differentiate similar functional groups in molecules. A list of protecting groups and how to introduce and remove these groups can be found in Greene, T.W., Wuts, P.G. M., *Protective Groups in Organic Synthesis*, 3<sup>rd</sup> edition, John Wiley & Sons: New York, 1999.

[027] It is to be understood that, unless otherwise stated, any description of a method of treatment or prevention includes use of the compounds to provide such treatment or prevention as is described herein. It is to be further understood, unless otherwise stated, any description of a method of treatment or prevention includes use of the compounds to prepare a medicament to treat or prevent such condition. The treatment or prevention includes treatment or prevention of human or non-human animals including rodents and other disease models.

[028] It is to be understood that, unless otherwise stated, any description of a method of treatment includes use of the compounds to provide such treatment or prophylaxis as is described herein, as well as use of the compounds to prepare a medicament to treat or prevent such condition. The treatment includes treatment of human or non-human animals including rodents and other disease models.

[029] As used herein, the term “subject” includes human and non-human animals, as well as cell lines, cell cultures, tissues, and organs. In some embodiments, the subject is a mammal. The mammal can be *e.g.*, a human or appropriate non-human mammal, such as primate, mouse, rat, dog, cat, cow, horse, goat, camel, sheep or a pig. The subject can also be a bird or fowl. In some embodiments, the subject is a human.

[030] As used herein, the term “subject in need thereof” refers to a subject having a disease or having an increased risk of developing the disease. A subject in need thereof can be one who has been previously diagnosed or identified as having a disease or disorder disclosed herein. A subject in need thereof can also be one who is suffering from a disease or disorder disclosed herein. Alternatively, a subject in need thereof can be one who has an increased risk of developing such disease or disorder relative to the population at large (*i.e.*, a subject who is predisposed to developing such disorder relative to the population at large). A subject in need thereof can have a refractory or resistant a disease or disorder disclosed herein (*i.e.*, a disease or disorder disclosed herein that does not respond or has not yet responded to treatment). The subject may be resistant at start of treatment or may become resistant during treatment. In some embodiments, the subject in need thereof received and failed all known effective therapies for a disease or disorder disclosed herein. In some embodiments, the subject in need thereof received at least one prior therapy.

[031] As used herein, the term “treating” or “treat” describes the management and care of a patient for the purpose of combating a disease, condition, or disorder and includes the administration of a compound of the present disclosure, or a pharmaceutically acceptable salt, polymorph or solvate thereof, to alleviate the symptoms or complications of a disease, condition or disorder, or to eliminate the disease, condition or disorder. The term “treat” can also include treatment of a cell *in vitro* or an animal model. It is to be appreciated that references to “treating” or “treatment” include the alleviation of established symptoms of a condition. “Treating” or “treatment” of a state, disorder or condition therefore includes: (1) preventing or delaying the appearance of clinical symptoms of the state, disorder or condition developing in a human that may be afflicted with or predisposed to the state, disorder or condition but does not yet experience or display clinical or subclinical symptoms of the state, disorder or condition, (2) inhibiting the state, disorder or condition, *i.e.*, arresting, reducing or delaying the development of the disease or a relapse thereof (in case of maintenance treatment) or at least one clinical or subclinical symptom thereof, or (3)

relieving or attenuating the disease, i.e., causing regression of the state, disorder or condition or at least one of its clinical or subclinical symptoms.

[032] It is to be understood that a compound of the present disclosure, or a pharmaceutically acceptable salt, polymorph or solvate thereof, can or may also be used to prevent a relevant disease, condition or disorder, or used to identify suitable candidates for such purposes.

[033] As used herein, the term “preventing,” “prevent,” or “protecting against” describes reducing or eliminating the onset of the symptoms or complications of such disease, condition or disorder.

[034] It is to be understood that one skilled in the art may refer to general reference texts for detailed descriptions of known techniques discussed herein or equivalent techniques. These texts include Ausubel *et al.*, *Current Protocols in Molecular Biology*, John Wiley and Sons, Inc. (2005); Sambrook *et al.*, *Molecular Cloning, A Laboratory Manual* (3<sup>rd</sup> edition), Cold Spring Harbor Press, Cold Spring Harbor, New York (2000); Coligan *et al.*, *Current Protocols in Immunology*, John Wiley & Sons, N.Y.; Enna *et al.*, *Current Protocols in Pharmacology*, John Wiley & Sons, N.Y.; Fingl *et al.*, *The Pharmacological Basis of Therapeutics* (1975), *Remington's Pharmaceutical Sciences*, Mack Publishing Co., Easton, PA, 18<sup>th</sup> edition (1990). These texts can, of course, also be referred to in making or using an aspect of the disclosure.

[035] It is to be understood that the present disclosure also provides pharmaceutical compositions comprising any compound described herein in combination with at least one pharmaceutically acceptable excipient or carrier.

[036] As used herein, the term “pharmaceutical composition” is a formulation containing the compounds of the present disclosure in a form suitable for administration to a subject. In one embodiment, the pharmaceutical composition is in bulk or in unit dosage form. The unit dosage form is any of a variety of forms, including, for example, a capsule, an IV bag, a tablet, a single pump on an aerosol inhaler or a vial. The quantity of active ingredient (*e.g.*, a formulation of the disclosed compound or salt, hydrate, solvate or isomer thereof) in a unit dose of composition is an effective amount and is varied according to the particular treatment involved. One skilled in the art will appreciate that it is sometimes necessary to make routine variations to the dosage depending on the age and condition of the patient. The dosage will also depend on the route of administration. A variety of routes are contemplated, including oral, pulmonary, rectal, parenteral, transdermal, subcutaneous, intravenous, intramuscular, intraperitoneal, inhalational, buccal, sublingual, intrapleural, intrathecal, intranasal, and the like. Dosage forms for the topical or

transdermal administration of a compound of this disclosure include powders, sprays, ointments, pastes, creams, lotions, gels, solutions, patches and inhalants. In one embodiment, the active compound is mixed under sterile conditions with a pharmaceutically acceptable carrier, and with any preservatives, buffers, or propellants that are required.

[037] As used herein, the term “pharmaceutically acceptable” refers to those compounds, anions, cations, materials, compositions, carriers, and/or dosage forms which are, within the scope of sound medical judgment, suitable for use in contact with the tissues of human beings and animals without excessive toxicity, irritation, allergic response, or other problem or complication, commensurate with a reasonable benefit/risk ratio.

[038] As used herein, the term “pharmaceutically acceptable excipient” means an excipient that is useful in preparing a pharmaceutical composition that is generally safe, non-toxic and neither biologically nor otherwise undesirable, and includes excipient that is acceptable for veterinary use as well as human pharmaceutical use. A “pharmaceutically acceptable excipient” as used in the specification and claims includes both one and more than one such excipient.

[039] It is to be understood that a pharmaceutical composition of the disclosure is formulated to be compatible with its intended route of administration. Examples of routes of administration include parenteral, *e.g.*, intravenous, intradermal, subcutaneous, oral (*e.g.*, ingestion), inhalation, transdermal (topical), and transmucosal administration. Solutions or suspensions used for parenteral, intradermal, or subcutaneous application can include the following components: a sterile diluent such as water for injection, saline solution, fixed oils, polyethylene glycols, glycerine, propylene glycol or other synthetic solvents; antibacterial agents such as benzyl alcohol or methyl parabens; antioxidants such as ascorbic acid or sodium bisulfite; chelating agents such as ethylenediaminetetraacetic acid; buffers such as acetates, citrates or phosphates, and agents for the adjustment of tonicity such as sodium chloride or dextrose. The pH can be adjusted with acids or bases, such as hydrochloric acid or sodium hydroxide. The parenteral preparation can be enclosed in ampoules, disposable syringes or multiple dose vials made of glass or plastic.

[040] It is to be understood that a compound or pharmaceutical composition of the disclosure can be administered to a subject in many of the well-known methods currently used for chemotherapeutic treatment. For example, a compound of the disclosure may be injected into the blood stream or body cavities or taken orally or applied through the skin with patches. The dose chosen should be sufficient to constitute effective treatment but not so high as to cause

unacceptable side effects. The state of the disease condition (*e.g.*, a disease or disorder disclosed herein) and the health of the patient should preferably be closely monitored during and for a reasonable period after treatment.

[041] As used herein, the term “therapeutically effective amount”, refers to an amount of a pharmaceutical agent to treat, ameliorate, or prevent an identified disease or condition, or to exhibit a detectable therapeutic or inhibitory effect. The effect can be detected by any assay method known in the art. The precise effective amount for a subject will depend upon the subject’s body weight, size, and health; the nature and extent of the condition; and the therapeutic or combination of therapeutics selected for administration. Therapeutically effective amounts for a given situation can be determined by routine experimentation that is within the skill and judgment of the clinician.

[042] As used herein, the term “therapeutically effective amount”, refers to an amount of a pharmaceutical agent to treat or ameliorate an identified disease or condition, or to exhibit a detectable therapeutic or inhibitory effect. The effect can be detected by any assay method known in the art. The precise effective amount for a subject will depend upon the subject’s body weight, size, and health; the nature and extent of the condition; and the therapeutic or combination of therapeutics selected for administration. Therapeutically effective amounts for a given situation can be determined by routine experimentation that is within the skill and judgment of the clinician.

[043] It is to be understood that, for any compound, the therapeutically effective amount can be estimated initially either in cell culture assays, *e.g.*, of neoplastic cells, or in animal models, usually rats, mice, rabbits, dogs, or pigs. The animal model may also be used to determine the appropriate concentration range and route of administration. Such information can then be used to determine useful doses and routes for administration in humans. Therapeutic/prophylactic efficacy and toxicity may be determined by standard pharmaceutical procedures in cell cultures or experimental animals, *e.g.*, ED<sub>50</sub> (the dose therapeutically effective in 50 % of the population) and LD<sub>50</sub> (the dose lethal to 50 % of the population). The dose ratio between toxic and therapeutic effects is the therapeutic index, and it can be expressed as the ratio, LD<sub>50</sub>/ED<sub>50</sub>. Pharmaceutical compositions that exhibit large therapeutic indices are preferred. The dosage may vary within this range depending upon the dosage form employed, sensitivity of the patient, and the route of administration.

[044] Dosage and administration are adjusted to provide sufficient levels of the active agent(s) or to maintain the desired effect. Factors which may be taken into account include the severity of the

disease state, general health of the subject, age, weight, and gender of the subject, diet, time and frequency of administration, drug combination(s), reaction sensitivities, and tolerance/response to therapy. Long-acting pharmaceutical compositions may be administered every 3 to 4 days, every week, or once every two weeks depending on half-life and clearance rate of the particular formulation.

[045] The pharmaceutical compositions containing active compounds of the present disclosure may be manufactured in a manner that is generally known, *e.g.*, by means of conventional mixing, dissolving, granulating, dragee-making, levigating, emulsifying, encapsulating, entrapping, or lyophilizing processes. Pharmaceutical compositions may be formulated in a conventional manner using one or more pharmaceutically acceptable carriers comprising excipients and/or auxiliaries that facilitate processing of the active compounds into preparations that can be used pharmaceutically. Of course, the appropriate formulation is dependent upon the route of administration chosen.

[046] Pharmaceutical compositions suitable for injectable use include sterile aqueous solutions (where water soluble) or dispersions and sterile powders for the extemporaneous preparation of sterile injectable solutions or dispersion. For intravenous administration, suitable carriers include physiological saline, bacteriostatic water, Cremophor EL™ (BASF, Parsippany, N.J.) or phosphate buffered saline (PBS). In all cases, the composition must be sterile and should be fluid to the extent that easy syringeability exists. It must be stable under the conditions of manufacture and storage and must be preserved against the contaminating action of microorganisms such as bacteria and fungi. The carrier can be a solvent or dispersion medium containing, for example, water, ethanol, polyol (for example, glycerol, propylene glycol, and liquid polyethylene glycol, and the like), and suitable mixtures thereof. The proper fluidity can be maintained, for example, by the use of a coating such as lecithin, by the maintenance of the required particle size in the case of dispersion and by the use of surfactants. Prevention of the action of microorganisms can be achieved by various antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, ascorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, polyalcohols such as mannitol and sorbitol, and sodium chloride in the composition. Prolonged absorption of the injectable compositions can be brought about by including in the composition an agent which delays absorption, for example, aluminum monostearate and gelatin.

[047] Sterile injectable solutions can be prepared by incorporating the active compound in the required amount in an appropriate solvent with one or a combination of ingredients enumerated above, as required, followed by filtered sterilization. Generally, dispersions are prepared by incorporating the active compound into a sterile vehicle that contains a basic dispersion medium and the required other ingredients from those enumerated above. In the case of sterile powders for the preparation of sterile injectable solutions, methods of preparation are vacuum drying and freeze-drying that yields a powder of the active ingredient plus any additional desired ingredient from a previously sterile-filtered solution thereof.

[048] Oral compositions generally include an inert diluent or an edible pharmaceutically acceptable carrier. They can be enclosed in gelatin capsules or compressed into tablets. For the purpose of oral therapeutic administration, the active compound can be incorporated with excipients and used in the form of tablets, troches, or capsules. Oral compositions can also be prepared using a fluid carrier for use as a mouthwash, wherein the compound in the fluid carrier is applied orally and swished and expectorated or swallowed. Pharmaceutically compatible binding agents, and/or adjuvant materials can be included as part of the composition. The tablets, pills, capsules, troches and the like can contain any of the following ingredients, or compounds of a similar nature: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a disintegrating agent such as alginic acid, Primogel, or corn starch; a lubricant such as magnesium stearate or Sterotes; a glidant such as colloidal silicon dioxide; a sweetening agent such as sucrose or saccharin; or a flavoring agent such as peppermint, methyl salicylate, or orange flavoring.

[049] For administration by inhalation, the compounds are delivered in the form of an aerosol spray from pressured container or dispenser, which contains a suitable propellant, *e.g.*, a gas such as carbon dioxide, or a nebulizer.

[050] Systemic administration can also be by transmucosal or transdermal means. For transmucosal or transdermal administration, penetrants appropriate to the barrier to be permeated are used in the formulation. Such penetrants are generally known in the art, and include, for example, for transmucosal administration, detergents, bile salts, and fusidic acid derivatives. Transmucosal administration can be accomplished through the use of nasal sprays or suppositories. For transdermal administration, the active compounds are formulated into ointments, salves, gels, or creams as generally known in the art.

[051] The active compounds can be prepared with pharmaceutically acceptable carriers that will protect the compound against rapid elimination from the body, such as a controlled release formulation, including implants and microencapsulated delivery systems. Biodegradable, biocompatible polymers can be used, such as ethylene vinyl acetate, polyanhydrides, polyglycolic acid, collagen, polyorthoesters, and polylactic acid. Methods for preparation of such formulations will be apparent to those skilled in the art. The materials can also be obtained commercially from Alza Corporation and Nova Pharmaceuticals, Inc. Liposomal suspensions (including liposomes targeted to infected cells with monoclonal antibodies to viral antigens) can also be used as pharmaceutically acceptable carriers. These can be prepared according to methods known to those skilled in the art, for example, as described in U.S. Pat. No. 4,522,811.

[052] It is especially advantageous to formulate oral or parenteral compositions in dosage unit form for ease of administration and uniformity of dosage. Dosage unit form as used herein refers to physically discrete units suited as unitary dosages for the subject to be treated; each unit containing a predetermined quantity of active compound calculated to produce the desired therapeutic effect in association with the required pharmaceutical carrier. The specification for the dosage unit forms of the disclosure are dictated by and directly dependent on the unique characteristics of the active compound and the particular therapeutic effect to be achieved.

[053] In therapeutic applications, the dosages of the pharmaceutical compositions used in accordance with the disclosure vary depending on the agent, the age, weight, and clinical condition of the recipient patient, and the experience and judgment of the clinician or practitioner administering the therapy, among other factors affecting the selected dosage. Generally, the dose should be sufficient to result in slowing, and preferably regressing, the symptoms of the disease or disorder disclosed herein and also preferably causing complete regression of the disease or disorder. Dosages can range from about 0.01 mg/kg per day to about 5000 mg/kg per day. An effective amount of a pharmaceutical agent is that which provides an objectively identifiable improvement as noted by the clinician or other qualified observer. Improvement in survival and growth indicates regression. As used herein, the term “dosage effective manner” refers to amount of an active compound to produce the desired biological effect in a subject or cell.

[054] It is to be understood that the pharmaceutical compositions can be included in a container, pack, or dispenser together with instructions for administration.

[055] It is to be understood that, for the compounds of the present disclosure being capable of further forming salts, all of these forms are also contemplated within the scope of the claimed disclosure.

[056] As used herein, the term “pharmaceutically acceptable salts” refer to derivatives of the compounds of the present disclosure wherein the parent compound is modified by making acid or base salts thereof. Examples of pharmaceutically acceptable salts include, but are not limited to, mineral or organic acid salts of basic residues such as amines, alkali or organic salts of acidic residues such as carboxylic acids, and the like. The pharmaceutically acceptable salts include the conventional non-toxic salts or the quaternary ammonium salts of the parent compound formed, for example, from non-toxic inorganic or organic acids. For example, such conventional non-toxic salts include, but are not limited to, those derived from inorganic and organic acids selected from 2-acetoxybenzoic, 2-hydroxyethane sulfonic, acetic, ascorbic, benzene sulfonic, benzoic, bicarbonic, carbonic, citric, edetic, ethane disulfonic, 1,2-ethane sulfonic, fumaric, glucoheptonic, gluconic, glutamic, glycolic, glycollyarsanilic, hexylresorcinic, hydrabamic, hydrobromic, hydrochloric, hydroiodic, hydroxymaleic, hydroxynaphthoic, isethionic, lactic, lactobionic, lauryl sulfonic, maleic, malic, mandelic, methane sulfonic, napsylic, nitric, oxalic, pamoic, pantothenic, phenylacetic, phosphoric, polygalacturonic, propionic, salicylic, stearic, subacetic, succinic, sulfamic, sulfanilic, sulfuric, tannic, tartaric, toluene sulfonic, and the commonly occurring amine acids, *e.g.*, glycine, alanine, phenylalanine, arginine, etc.

[057] In some embodiments, the pharmaceutically acceptable salt is a sodium salt, a potassium salt, a calcium salt, a magnesium salt, a diethylamine salt, a choline salt, a meglumine salt, a benzathine salt, a tromethamine salt, an ammonia salt, an arginine salt, or a lysine salt.

[058] Other examples of pharmaceutically acceptable salts include hexanoic acid, cyclopentane propionic acid, pyruvic acid, malonic acid, 3-(4-hydroxybenzoyl)benzoic acid, cinnamic acid, 4-chlorobenzenesulfonic acid, 2-naphthalenesulfonic acid, 4-toluenesulfonic acid, camphorsulfonic acid, 4-methylbicyclo-[2.2.2]-oct-2-ene-1-carboxylic acid, 3-phenylpropionic acid, trimethylacetic acid, tertiary butylacetic acid, muconic acid, and the like. The present disclosure also encompasses salts formed when an acidic proton present in the parent compound either is replaced by a metal ion, *e.g.*, an alkali metal ion, an alkaline earth ion, or an aluminum ion; or coordinates with an organic base such as ethanolamine, diethanolamine, triethanolamine, tromethamine, N-methylglucamine, and the like. In the salt form, it is understood that the ratio of

the compound to the cation or anion of the salt can be 1:1, or any ratio other than 1:1, e.g., 3:1, 2:1, 1:2, or 1:3.

[059] It is to be understood that all references to pharmaceutically acceptable salts include solvent addition forms (solvates) or crystal forms (polymorphs) as defined herein, of the same salt.

[060] The compounds, or pharmaceutically acceptable salts thereof, are administered orally, nasally, transdermally, pulmonary, inhalationally, buccally, sublingually, intraperitoneally, subcutaneously, intramuscularly, intravenously, rectally, intrapleurally, intrathecally and parenterally. In one embodiment, the compound is administered orally. One skilled in the art will recognize the advantages of certain routes of administration.

[061] The dosage regimen utilizing the compounds is selected in accordance with a variety of factors including type, species, age, weight, sex and medical condition of the patient; the severity of the condition to be treated; the route of administration; the renal and hepatic function of the patient; and the particular compound or salt thereof employed. An ordinarily skilled physician or veterinarian can readily determine and prescribe the effective amount of the drug required to prevent, counter, or arrest the progress of the condition. An ordinarily skilled physician or veterinarian can readily determine and prescribe the effective amount of the drug required to counter or arrest the progress of the condition.

[062] Techniques for formulation and administration of the disclosed compounds of the disclosure can be found in *Remington: the Science and Practice of Pharmacy*, 19<sup>th</sup> edition, Mack Publishing Co., Easton, PA (1995). In an embodiment, the compounds described herein, and the pharmaceutically acceptable salts thereof, are used in pharmaceutical preparations in combination with a pharmaceutically acceptable carrier or diluent. Suitable pharmaceutically acceptable carriers include inert solid fillers or diluents and sterile aqueous or organic solutions. The compounds will be present in such pharmaceutical compositions in amounts sufficient to provide the desired dosage amount in the range described herein.

[063] All percentages and ratios used herein, unless otherwise indicated, are by weight. Other features and advantages of the present disclosure are apparent from the different examples. The provided examples illustrate different components and methodology useful in practicing the present disclosure. The examples do not limit the claimed disclosure. Based on the present disclosure the skilled artisan can identify and employ other components and methodology useful for practicing the present disclosure.

[064] In the synthetic scheme described herein, compounds may be drawn with one particular configuration for simplicity. Such particular configurations are not to be construed as limiting the disclosure to one or another isomer, tautomer, regioisomer or stereoisomer, nor does it exclude mixtures of isomers, tautomers, regioisomers or stereoisomers; however, it will be understood that a given isomer, tautomer, regioisomer or stereoisomer may have a higher level of activity than another isomer, tautomer, regioisomer or stereoisomer.

[065] All publications and patent documents cited herein are incorporated herein by reference as if each such publication or document was specifically and individually indicated to be incorporated herein by reference. Citation of publications and patent documents is not intended as an admission that any is pertinent prior art, nor does it constitute any admission as to the contents or date of the same. The invention having now been described by way of written description, those of skill in the art will recognize that the invention can be practiced in a variety of embodiments and that the foregoing description and examples below are for purposes of illustration and not limitation of the claims that follow.

[066] As use herein, the phrase “compound of the disclosure” refers to those compounds which are disclosed herein, both generically and specifically.

### *Biological Assays*

[067] Compound A, or a pharmaceutically acceptable salt thereof, can be characterized using a variety of assays known to those skilled in the art to determine whether Compound A has biological activity. For example, Compound A can be characterized by conventional assays, including but not limited to those assays described below, to determine whether they have a predicted activity, binding activity and/or binding specificity.

[068] Furthermore, high-throughput screening can be used to speed up analysis using such assays. As a result, it can be possible to rapidly screen the molecule described herein for activity, using techniques known in the art. General methodologies for performing high-throughput screening are described, for example, in Devlin (1998) *High Throughput Screening*, Marcel Dekker; and U.S. Patent No. 5,763,263. High-throughput assays can use one or more different assay techniques including, but not limited to, those described below.

[069] Various *in vitro* or *in vivo* biological assays are may be suitable for detecting the effect of Compound A, or a pharmaceutically acceptable salt thereof. These *in vitro* or *in vivo* biological

assays can include, but are not limited to, enzymatic activity assays, electrophoretic mobility shift assays, reporter gene assays, *in vitro* cell viability assays, and the assays described herein.

[070] In some embodiments, for irritable bowel syndrome with constipation, the defecation component may be evaluated by assessing stool frequency. Stool frequency, as measured by the number of complete spontaneous bowel movements (CSBMs) per week may be useful in defining a treatment response in chronic constipation for irritable bowel syndrome with constipation patients.

[071] In some embodiments, for irritable bowel syndrome with diarrhea, the defecation component may be evaluated by assessing stool consistency. The Bristol Stool Form Scale may provide a description of stool consistency and form and may be an appropriate guide for capturing stool consistency in irritable bowel syndrome trials.

[072] In some embodiments, a colonic sensitive assessment may be used to assess colonic hypersensitivity (e.g., acute acetic acid-induced colonic hypersensitivity) in a subject (e.g., a rat) may be put under a brief anesthesia, for colonic sensitivity assessment. A visceromotor response (VMR) to colorectal distension (CRD) may be used to assess colonic sensitivity. The subject (e.g., rat) may receive an intracolonic (i.c.) administration of Compound A or vehicle. Then, terminal blood may be collected and blood plasma isolated for assessment.

[073] In some embodiments, Wrap Restraint Stress Model may be used to induces an IBS phenotype in a subject (e.g., rat). Measurement of colorectal distension in rats at different balloon volumes may produce abdominal contractions (e.g., spasms or cramps) due to pain. The effects of Compound A may be evaluated with a wrap restraint stress model. A standard of care for IBS (e.g., Eluxadoline, and morphine) may be used control compounds for a wrap restraint stress model.

[074] In some embodiments, the biological assay is described in the Examples herein.

### *Pharmaceutical Compositions*

[075] In some aspects, the present disclosure provides a pharmaceutical composition comprising Compound A as an active ingredient. In some embodiments, the present disclosure provides a pharmaceutical composition comprising Compound A, or a pharmaceutically acceptable salt thereof, and one or more pharmaceutically acceptable carriers or excipients.

[076] As used herein, the term “composition” is intended to encompass a product comprising the specified ingredients in the specified amounts, as well as any product which results, directly or indirectly, from combination of the specified ingredients in the specified amounts.

[077] Compound A can be formulated for oral administration in forms such as tablets, capsules (each of which includes sustained release or timed release formulations), pills, powders, granules, elixirs, tinctures, suspensions, syrups and emulsions. Compound A can also be formulated for intravenous (bolus or in-fusion), intraperitoneal, topical, subcutaneous, intramuscular or transdermal (*e.g.*, patch) administration, all using forms well known to those of ordinary skill in the pharmaceutical arts.

[078] The formulation of the present disclosure may be in the form of an aqueous solution comprising an aqueous vehicle. The aqueous vehicle component may comprise water and at least one pharmaceutically acceptable excipient. Suitable acceptable excipients include those selected from the group consisting of a solubility enhancing agent, chelating agent, preservative, tonicity agent, viscosity/suspending agent, buffer, and pH modifying agent, and a mixture thereof.

[079] Any suitable solubility enhancing agent can be used. Examples of a solubility enhancing agent include cyclodextrin, such as those selected from the group consisting of hydroxypropyl- $\beta$ -cyclodextrin, methyl- $\beta$ -cyclodextrin, randomly methylated- $\beta$ -cyclodextrin, ethylated- $\beta$ -cyclodextrin, triacetyl- $\beta$ -cyclodextrin, peracetylated- $\beta$ -cyclodextrin, carboxymethyl- $\beta$ -cyclodextrin, hydroxyethyl- $\beta$ -cyclodextrin, 2-hydroxy-3-(trimethylammonio)propyl- $\beta$ -cyclodextrin, glucosyl- $\beta$ -cyclodextrin, sulfated  $\beta$ -cyclodextrin (S- $\beta$ -CD), maltosyl- $\beta$ -cyclodextrin,  $\beta$ -cyclodextrin sulfobutyl ether, branched- $\beta$ -cyclodextrin, hydroxypropyl- $\gamma$ -cyclodextrin, randomly methylated- $\gamma$ -cyclodextrin, and trimethyl- $\gamma$ -cyclodextrin, and mixtures thereof.

[080] Any suitable chelating agent can be used. Examples of a suitable chelating agent include those selected from the group consisting of ethylenediaminetetraacetic acid and metal salts thereof, disodium edetate, trisodium edetate, and tetrasodium edetate, and mixtures thereof.

[081] Any suitable preservative can be used. Examples of a preservative include those selected from the group consisting of quaternary ammonium salts such as benzalkonium halides (preferably benzalkonium chloride), chlorhexidine gluconate, benzethonium chloride, cetyl pyridinium chloride, benzyl bromide, phenylmercury nitrate, phenylmercury acetate, phenylmercury neodecanoate, merthiolate, methylparaben, propylparaben, sorbic acid, potassium sorbate, sodium

benzoate, sodium propionate, ethyl p-hydroxybenzoate, propylaminopropyl biguanide, and butyl-p-hydroxybenzoate, and sorbic acid, and mixtures thereof.

[082] The aqueous vehicle may also include a tonicity agent to adjust the tonicity (osmotic pressure). The tonicity agent can be selected from the group consisting of a glycol (such as propylene glycol, diethylene glycol, triethylene glycol), glycerol, dextrose, glycerin, mannitol, potassium chloride, and sodium chloride, and a mixture thereof.

[083] The aqueous vehicle may also contain a viscosity/suspending agent. Suitable viscosity/suspending agents include those selected from the group consisting of cellulose derivatives, such as methyl cellulose, ethyl cellulose, hydroxyethylcellulose, polyethylene glycols (such as polyethylene glycol 300, polyethylene glycol 400), carboxymethyl cellulose, hydroxypropylmethyl cellulose, and cross-linked acrylic acid polymers (carbomers), such as polymers of acrylic acid cross-linked with polyalkenyl ethers or divinyl glycol (Carbopols - such as Carbopol 934, Carbopol 934P, Carbopol 971, Carbopol 974 and Carbopol 974P), and a mixture thereof.

[084] In order to adjust the formulation to an acceptable pH (typically a pH range of about 5.0 to about 9.0, more preferably about 5.5 to about 8.5, particularly about 6.0 to about 8.5, about 7.0 to about 8.5, about 7.2 to about 7.7, about 7.1 to about 7.9, or about 7.5 to about 8.0), the formulation may contain a pH modifying agent. The pH modifying agent is typically a mineral acid or metal hydroxide base, selected from the group of potassium hydroxide, sodium hydroxide, and hydrochloric acid, and mixtures thereof, and preferably sodium hydroxide and/or hydrochloric acid. These acidic and/or basic pH modifying agents are added to adjust the formulation to the target acceptable pH range. Hence it may not be necessary to use both acid and base - depending on the formulation, the addition of one of the acid or base may be sufficient to bring the mixture to the desired pH range.

[085] The aqueous vehicle may also contain a buffering agent to stabilize the pH. When used, the buffer is selected from the group consisting of a phosphate buffer (such as sodium dihydrogen phosphate and disodium hydrogen phosphate), a borate buffer (such as boric acid, or salts thereof including disodium tetraborate), a citrate buffer (such as citric acid, or salts thereof including sodium citrate), and  $\epsilon$ -aminocaproic acid, and mixtures thereof.

[086] The formulation may further comprise a wetting agent. Suitable classes of wetting agents include those selected from the group consisting of polyoxypropylene-polyoxyethylene block

copolymers (poloxamers), polyethoxylated ethers of castor oils, polyoxyethylenated sorbitan esters (polysorbates), polymers of oxyethylated octyl phenol (Tyloxapol), polyoxyl 40 stearate, fatty acid glycol esters, fatty acid glyceryl esters, sucrose fatty esters, and polyoxyethylene fatty esters, and mixtures thereof.

[087] Oral compositions generally include an inert diluent or an edible pharmaceutically acceptable carrier. They can be enclosed in gelatin capsules or compressed into tablets. For the purpose of oral therapeutic administration, the active compound can be incorporated with excipients and used in the form of tablets, troches, or capsules. Oral compositions can also be prepared using a fluid carrier for use as a mouthwash, wherein the compound in the fluid carrier is applied orally and swished and expectorated or swallowed. Pharmaceutically compatible binding agents, and/or adjuvant materials can be included as part of the composition. The tablets, pills, capsules, troches and the like can contain any of the following ingredients, or compounds of a similar nature: a binder such as microcrystalline cellulose, gum tragacanth or gelatin; an excipient such as starch or lactose, a disintegrating agent such as alginic acid, Primogel, or corn starch; a lubricant such as magnesium stearate or Sterotes; a glidant such as colloidal silicon dioxide; a sweetening agent such as sucrose or saccharin; or a flavoring agent such as peppermint, methyl salicylate, or orange flavoring.

[088] According to a further aspect of the disclosure there is provided a pharmaceutical composition which comprises a compound of the disclosure as defined hereinbefore, or a pharmaceutically acceptable salt, hydrate or solvate thereof, in association with a pharmaceutically acceptable diluent or carrier.

[089] The compositions of the disclosure may be in a form suitable for oral use (for example as tablets, lozenges, hard or soft capsules, aqueous or oily suspensions, emulsions, dispersible powders or granules, syrups or elixirs), for topical use (for example as creams, ointments, gels, or aqueous or oily solutions or suspensions), for administration by inhalation (for example as a finely divided powder or a liquid aerosol), for administration by insufflation (for example as a finely divided powder) or for parenteral administration (for example as a sterile aqueous or oily solution for intravenous, subcutaneous, intramuscular, intraperitoneal or intramuscular dosing or as a suppository for rectal dosing).

[090] The compositions of the disclosure may be obtained by conventional procedures using conventional pharmaceutical excipients, well known in the art. Thus, compositions intended for

oral use may contain, for example, one or more coloring, sweetening, flavoring and/or preservative agents.

[091] An effective amount of Compound A for use in therapy is an amount sufficient to treat or prevent irritable bowel syndrome, slow its progression and/or reduce the symptoms associated with the condition.

[092] An effective amount of Compound A for use in therapy is an amount sufficient to treat irritable bowel syndrome, slow its progression and/or reduce the symptoms associated with the condition.

[093] The size of the dose for therapeutic or prophylactic purposes of Compound A will naturally vary according to the nature and severity of the conditions, the age and sex of the animal or patient and the route of administration, according to well-known principles of medicine.

[094] Examples of useful dermatological compositions which can be used to deliver Compound A to the skin are known to the art; for example, see Jacquet et al. (U.S. Pat. No. 4,608,392), Geria (U.S. Pat. No. 4,992,478), Smith et al. (U.S. Pat. No. 4,559,157) and Wortzman (U.S. Pat. No. 4,820,508).

#### *Methods of Use*

[095] In some aspects, the present disclosure provides a method of treating or preventing a disease or disorder disclosed herein in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[096] In some aspects, the present disclosure provides a method of treating a disease or disorder disclosed herein in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[097] In some aspects, the present disclosure provides a method of treating or preventing a disease or disorder disclosed herein in a subject in need thereof, comprising administering to the subject Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[098] In some aspects, the present disclosure provides a method of treating a disease or disorder disclosed herein in a subject in need thereof, comprising administering to the subject Compound

A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0099] In some embodiments, the disease or disorder is irritable bowel syndrome.

[0100] In some aspects, the present disclosure provides a method of reducing colonic hypersensitivity in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0101] In some aspects, the present disclosure provides a method of reducing colonic hypersensitivity in a subject in need thereof, comprising administering to the subject Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0102] In some aspects, the present disclosure provides a method of treating or preventing irritable bowel syndrome in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0103] In some aspects, the present disclosure provides a method of treating irritable bowel syndrome in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0104] In some aspects, the present disclosure provides a method of treating or preventing irritable bowel syndrome in a subject in need thereof, comprising administering to the subject Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0105] In some aspects, the present disclosure provides a method of treating irritable bowel syndrome in a subject in need thereof, comprising administering to the subject Compound A or a pharmaceutically acceptable salt thereof, or a pharmaceutical composition of the present disclosure.

[0106] In some aspects, the present disclosure provides Compound A or a pharmaceutically acceptable salt thereof for use in treating or preventing a disease or disorder disclosed herein in a subject in need thereof.

[0107] In some aspects, the present disclosure provides Compound A or a pharmaceutically acceptable salt thereof for use in treating a disease or disorder disclosed herein in a subject in need thereof.

[0108] In some aspects, the present disclosure provides Compound A or a pharmaceutically acceptable salt thereof for use in reducing colonic hypersensitivity in a subject in need thereof.

[0109] In some aspects, the present disclosure provides Compound A or a pharmaceutically acceptable salt thereof for use in treating or preventing irritable bowel syndrome in a subject in need thereof.

[0110] In some aspects, the present disclosure provides Compound A or a pharmaceutically acceptable salt thereof for use in treating irritable bowel syndrome in a subject in need thereof.

[0111] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof in the manufacture of a medicament for treating or preventing a disease or disorder disclosed herein.

[0112] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof in the manufacture of a medicament for treating a disease or disorder disclosed herein.

[0113] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof in the manufacture of a medicament for reducing colonic hypersensitivity in a subject in need thereof.

[0114] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof in the manufacture of a medicament for treating or preventing irritable bowel syndrome in a subject in need thereof.

[0115] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof in the manufacture of a medicament for treating irritable bowel syndrome in a subject in need thereof.

[0116] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof for the treatment or prevention of a disease or disorder disclosed herein.

[0117] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof for the treatment of a disease or disorder disclosed herein.

[0118] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof for the reduction of colonic hypersensitivity.

[0119] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof for the treatment or prevention of irritable bowel syndrome.

[0120] In some aspects, the present disclosure provides use of Compound A or a pharmaceutically acceptable salt thereof for the treatment of irritable bowel syndrome.

[0121] Effectiveness of compounds of the disclosure can be determined by industry-accepted assays/ disease models according to standard practices of elucidating the same as described in the art and are found in the current general knowledge.

[0122] In some embodiments, the disease or disorder is irritable bowel syndrome.

[0123] In some embodiments, the irritable bowel syndrome is irritable bowel syndrome with diarrhea (IBS-D).

[0124] In some embodiments, the irritable bowel syndrome is irritable bowel syndrome with constipation (IBS-C).

[0125] In some embodiments, the disease or disorder is pain associated with irritable bowel syndrome.

[0126] In some embodiments, the disease or disorder is a symptom of irritable bowel syndrome.

[0127] In some embodiments, a symptom of irritable bowel syndrome is pain.

[0128] In some embodiments, the pain is abdominal pain. In some embodiments, the pain is discomfort.

[0129] In some embodiments, a symptom of irritable bowel syndrome is discomfort.

[0130] In some embodiments, a symptom of irritable bowel syndrome is altered bowel habit (e.g., stool frequency).

[0131] In some embodiments, the subject is a mammal. In some embodiments the mammal is a human.

[0132] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.5 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 1 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 2 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 3 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 4 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof,

is administered from about 5 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 6 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 7 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 8 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 9 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 10 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 15 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 20 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 25 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 30 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 35 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 40 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 45 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 50 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 75 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 100 mg to about 250 mg.

[0133] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 250 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 100 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 75 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 50 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 45 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 40 mg. In some embodiments, Compound A, or a

pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 40 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 35 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 30 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 25 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 20 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 15 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 10 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 9 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 8 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 7 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 6 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 5 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 4 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 3 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 2 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 1 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 0.5 mg.

[0134] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.1 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.5 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 1 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 2 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 3 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is

administered at about 4 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 5 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 6 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 7 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 8 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 9 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 10 mg.

[0135] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 15 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 20 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 25 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 30 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 35 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 40 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 45 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 50 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 75 mg.

[0136] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 100 mg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 250 mg.

[0137] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.025 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.050 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.075 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.100 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.150 mg/kg to about 30

mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.200 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.300 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.500 mg/kg to about 30 mg/kg.

[0138] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 1 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 1.5 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 2 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 2.5 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 3 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 3.5 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 4 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 4.5 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 5 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 10 mg/kg to about 30 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 20 mg/kg to about 30 mg/kg.

[0139] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 20 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 10 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 4.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 4 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 3.5

mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 3 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 2.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 2 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 1.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 1 mg/kg.

[0140] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.500 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.300 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.200 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.150 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.100 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.075 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.050 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 0.025 mg/kg.

[0141] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.010 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.025 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.050 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.075 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.100 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.150 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.200 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about

0.300 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 0.500 mg/kg.

[0142] In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 1 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 1.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 2 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 2.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 3 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 3.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 4 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 4.5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 5 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 10 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 20 mg/kg. In some embodiments, Compound A, or a pharmaceutically acceptable salt thereof, is administered at about 30 mg/kg.

[0143] In some embodiments, Compound A is administered once, twice, three times, four times, or five times per day. In some embodiments, Compound A is administered once daily. In some embodiments, Compound A is administered twice daily. In some embodiments, Compound A is administered three times daily. In some embodiments, Compound A is administered four times daily. In some embodiments, Compound A is administered five times daily.

[0144] In some embodiments, Compound A is administered with a drug holiday. In some embodiments, Compound A is administered without a drug holiday.

[0145] In some embodiments, Compound A is administered intracolonicallly. In some embodiments, Compound A is administered orally.

## EXAMPLES

### **Example 1. Acute Acetic Acid-Induced Colonic Hypersensitivity**

[0146] *Animals*: Male Sprague Dawley rats (300-350g) were purchased from Charles River Laboratories. Rats were acclimated to the laboratory and animal handler for 2 weeks.

[0147] *Induction of Colonic Hypersensitivity*: Under brief isoflurane anesthesia, acetic acid at 0.6% was infused into the colon (at a volume of 1.5 ml) 1-hr before colonic sensitivity assessment.

[0148] *Colonic Sensitivity Assessment*: Visceromotor response (VMR) to colorectal distension (CRD) was used to assess colonic sensitivity. The VMR was quantified as the number of abdominal contractions in response to 10 min CRD at randomized, graded isobaric pressures of 0, 20, 40 and 60 mmHg.

[0149] *Dosage*: Rats received intracolonic (i.c.) administration of Compound A (100µg/kg) or vehicle (0.5% methylcellulose), 30-min prior to measuring colonic sensitivity.

[0150] *Tissue Collection*: Terminal blood was collected and blood plasma was isolated and stored at -80°C. Following colonic hypersensitivity assessment, two 1-cm segments of colon were collected and stored accordingly:

- One section flash frozen in liquid nitrogen and stored in -80°C.
- One section fixed in 10% neutral buffered formalin for 24-hrs. and stored in 70% ethanol at room temperature.

[0151] *Statistical Analysis and Experimental Rigor*: All data was expressed as mean ± standard error of the mean (S.E.M.). Experimental groups were not randomized. Analyses were performed using GraphPad Prism 8 (GraphPad Software, USA). Statistical significance was determined using Two-Way Repeated Measure ANOVA followed by Bonferroni's multi-comparison test. A  $p < 0.05$  was considered statistically significant in all tests. The experimenter was not blinded to drug treatment.

[0152] FIG. 1A and 1B depict the effect of locally administered Compound A on acetic acid-induced colonic hypersensitivity, wherein A.A. denotes Acetic Acid.

## **Example 2. Wrap Restraint Stress Model**

[0153] Wrap restraint may cause neurological stress which induces an IBS phenotype in rats. Measurement of colorectal distension in rats at different balloon volumes may produce abdominal contractions (spasms/cramps) due to pain. The effects of Compound A at three doses (0.1, 1 and 10 mg/kg, intracolonic administration) in the rat model of Irritable Bowel Syndrome (IBS) induced

by wrap restraint stress (WRS) was tested. Standard of care for IBS, Eluxadoline, and morphine were used as control compounds. FIG. 2 depicts the wrap restraint stress model.

[0154] Complete reduction in abdominal contractions at 10 mg/kg of Compound A was observed (FIG. 3). The ED<sub>50</sub> is approximately 0.1 mg/kg and >90% efficacy at 1mg/kg. All doses were administered intracolonicallly. Morphine (4 mg/kg SC) also produced complete reversal of abdominal contractions, while the standard of care for IBS-D, Eluxadoline (10 mg/kg, PO), did not produce any significant reduction.

### **Example 3. Mu-Kappa GI Immunofluorescence**

[0155] Rat GI tissues from 3 male and 3 females were trimmed and processed. Each Formalin fixed paraffin embedded (FFPE) Sprague Dawley rat tissue was sectioned at 4µm onto positively charged slides for immunostaining. 2 plex Immunofluorescence (IF) was performed on FFPE rat colon tissue with a Leica Bond automated immunostainer and a rabbit anti-Mu (Neuromics, #RA10104) and a mouse anti-Kappa (Neuromics, #MO15098) opioid receptor antibody.

[0156] Heat induced antigen retrieval was performed using Leica Bond Epitope Retrieval Buffer 2 (EDTA solution, pH 9.0) for 20 minutes (ER2(20)). Non-specific antibody binding was blocked using Novolink Protein Block (Leica, cat#RE7280-CE) for 30 minutes. Primary antibodies were applied for overnight incubation at 4°C. Donkey anti-rabbit IgG Alexa Fluor® 546 (ThermoFisher, Cat# A10040, Lot# 2020130) and Donkey anti-mouse IgG Alexa Fluor® 647 (ThermoFisher, Cat#A31571, Lot#2136787) were applied for 60 minutes.

[0157] FIG. 4 shows a DAPI nuclear counterstain (blue) applied to the rat colon tissue. FIG. 4 shows strong co-expression of both mu and kappa opioid receptors (orange) in rat colon tissue. Several microvilli cells in the colon showed strong co-expression of mu-kappa opioid receptors. In view of both the targeting of the mu-kappa opioid receptors and anatomical region relevant to the IBS (*i.e.*, colon), an effective route for the treatment of IBS with Compound A (a potent and selective activator of mu-kappa opioid receptors) is shown. The activity exhibited in Examples 1 and 2 above (*i.e.*, that Compound A provides analgesic efficacy in IBS animal models) in combination with strong co-expression of mu-kappa opioid receptors of the colon, provides a route for determination that Compound A is a potent analgesic for the treatment of IBS.

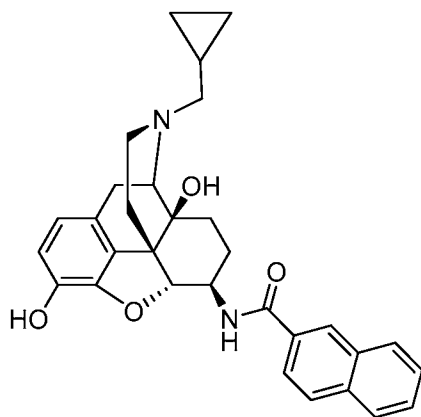
### EQUIVALENTS

[0158] The details of one or more embodiments of the disclosure are set forth in the accompanying description above. Although any methods and materials similar or equivalent to those described herein can be used in the practice or testing of the present disclosure, the preferred methods and materials are now described. Other features, objects, and advantages of the disclosure will be apparent from the description and from the claims. In the specification and the appended claims, the singular forms include plural referents unless the context clearly dictates otherwise. Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this disclosure belongs. All patents and publications cited in this specification are incorporated by reference.

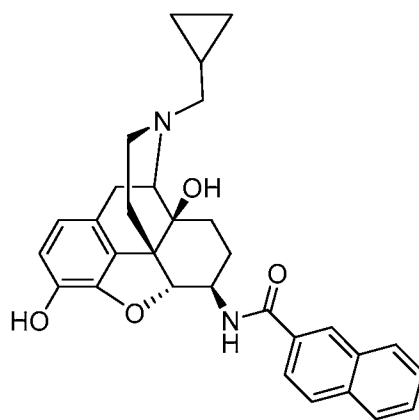
[0159] The foregoing description has been presented only for the purposes of illustration and is not intended to limit the disclosure to the precise form disclosed, but by the claims appended hereto.

**What is claimed is:**

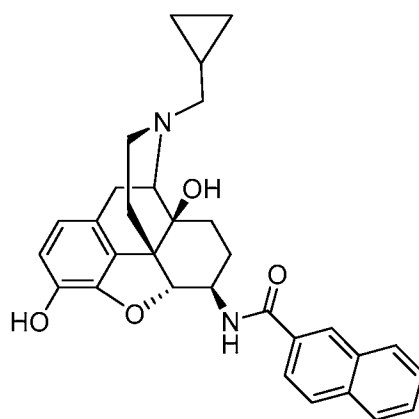
1. A method of treating a disease or disorder in a subject comprising administering to the



subject Compound A, (A), or a pharmaceutically acceptable salt thereof.

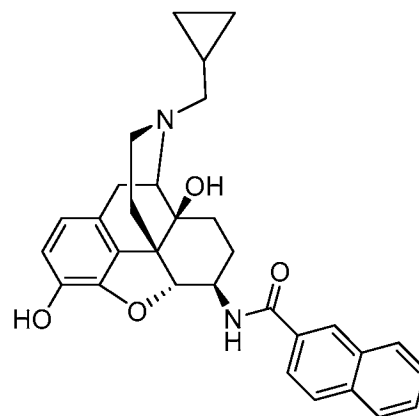


2. A compound having the structure of Compound A, or a pharmaceutically acceptable salt thereof, for use in treating a disease or disorder in a subject.



3. A compound having the structure of Compound A, or a pharmaceutically acceptable salt thereof, for use in the manufacture of a medicament for

treating a disease or disorder in a subject.



4. Use of a compound having the structure of Compound A, (A), or a pharmaceutically acceptable salt thereof, for treatment of a disease or disorder.
5. The method, use, or compound of any one of the preceding claims, wherein the disease or disorder is irritable bowel syndrome.
6. The method, use, or compound of any one of the preceding claims, wherein the irritable bowel syndrome is irritable bowel syndrome with diarrhea or irritable bowel syndrome with constipation.
7. The method, use, or compound of any one of the preceding claims, wherein the disease or disorder is pain associated with irritable bowel syndrome.
8. The method, use, or compound of any one of the preceding claims, wherein the disease or disorder is a symptom of irritable bowel syndrome.
9. The method, use, or compound of claim 8, wherein the symptom of irritable bowel syndrome is pain.
10. The method, use, or compound of claim 9, wherein the pain is abdominal pain or discomfort.

11. The method, use, or compound of claim 8, wherein the of irritable bowel syndrome is altered bowel habit (e.g., stool frequency).
12. The method, use, or compound of any one of the preceding claims, wherein the subject is human.
13. The method, use, or compound of any one of the preceding claims, wherein Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.1 mg to about 250 mg.
14. The method, use, or compound of any one of the preceding claims, wherein Compound A, or a pharmaceutically acceptable salt thereof, is administered from about 0.010 mg/kg to about 30 mg/kg.
15. The method, use, or compound of any one of the preceding claims, wherein Compound A is administered once, twice, three times, four times, or five times per day.

FIG. 1A

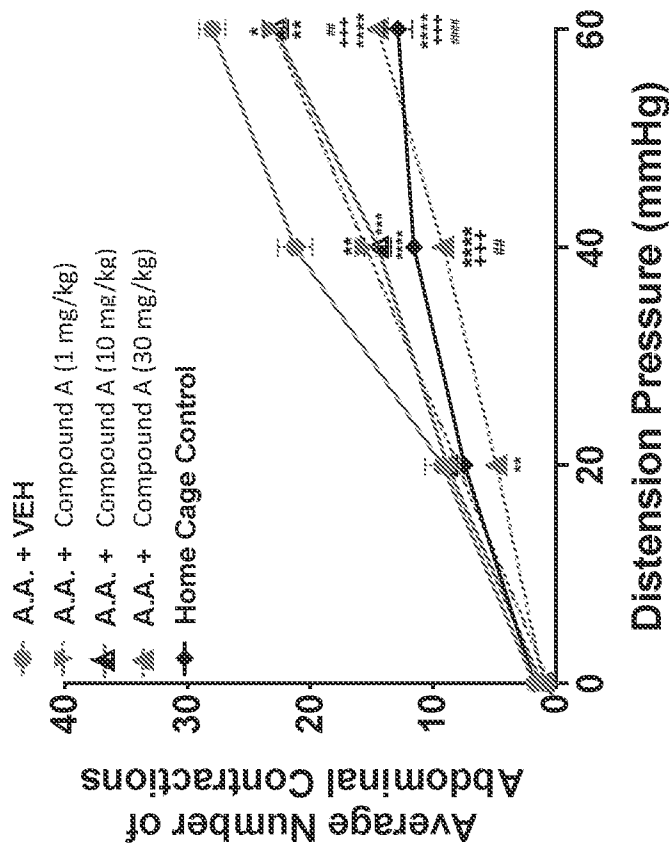
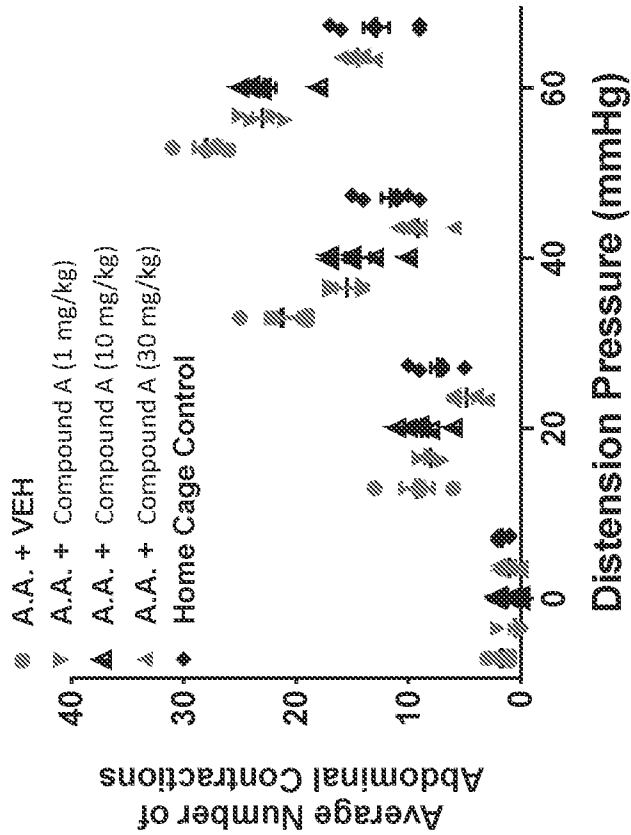
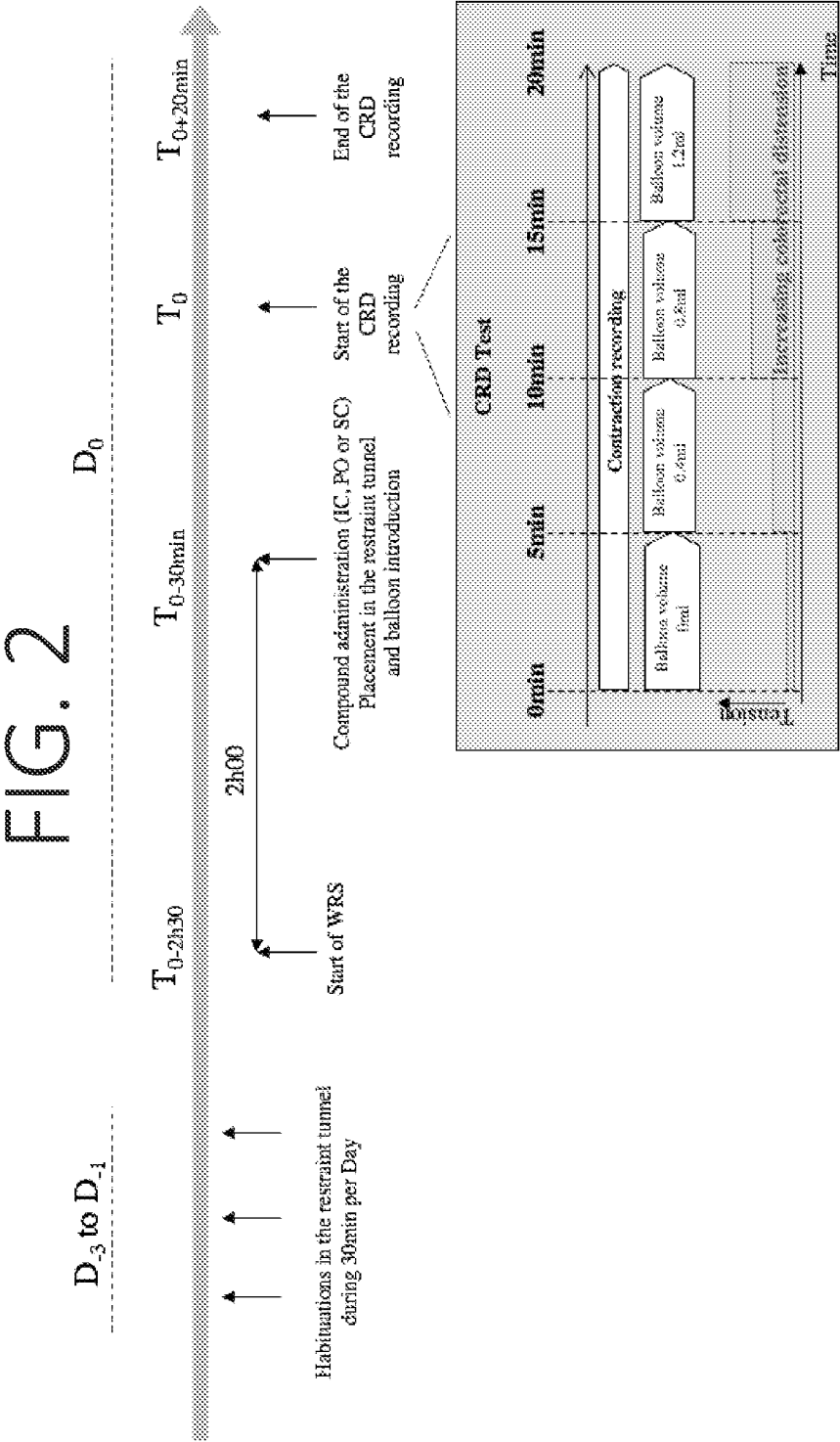


FIG. 1B



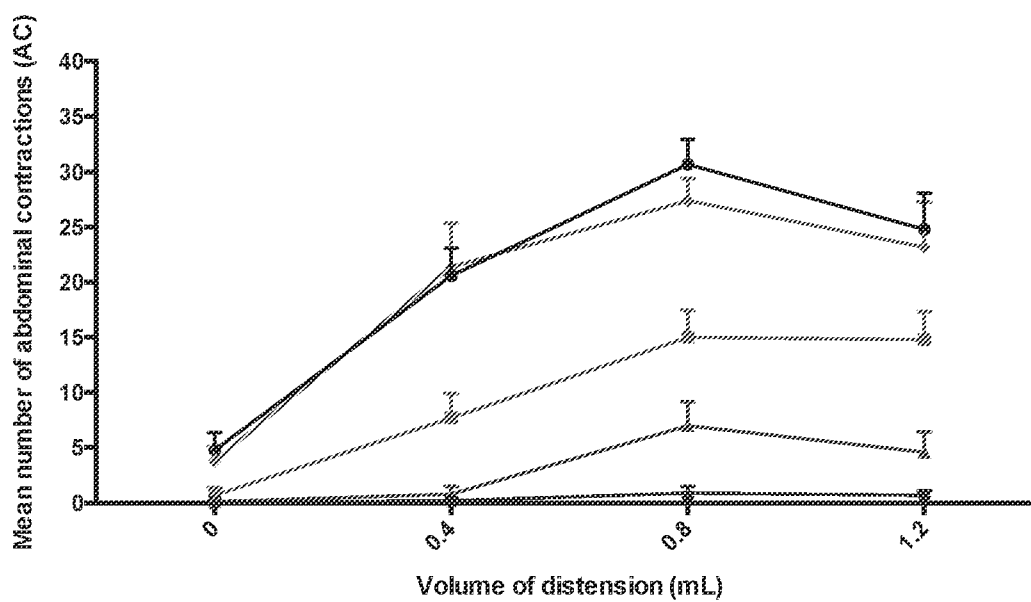
\*\*\*p<0.01 \*\*\*\*p<0.0001 compared to A.A. + VEH, \*\*\*p<0.001, \*\*\*\*p<0.0001 compared to A.A. + Compound A (1mg/kg).  
####p<0.0001 compared to A.A. + Compound A (100µg/kg), RM Two-Way ANOVA/ Bonferroni multi-comparison post-test.



CRD, colorectal distension test; D, Day; h, hour; IC, intracolonic; min, minute; ml, milliliter; PO, per os; SC, subcutaneous; T, time; WRS, wrap restraint stress

## FIG. 3

Data on CRD following WRS sensitization



Group 1\_Placebo      Group 4\_Compound A (10 mg/kg)  
Group 2\_Compound A (0.1 mg/kg)      Group 5\_Eluxadoline 10mg/Kg  
Group 3\_Compound A (1 mg/kg)      Group 6\_Morphine 4mg/Kg

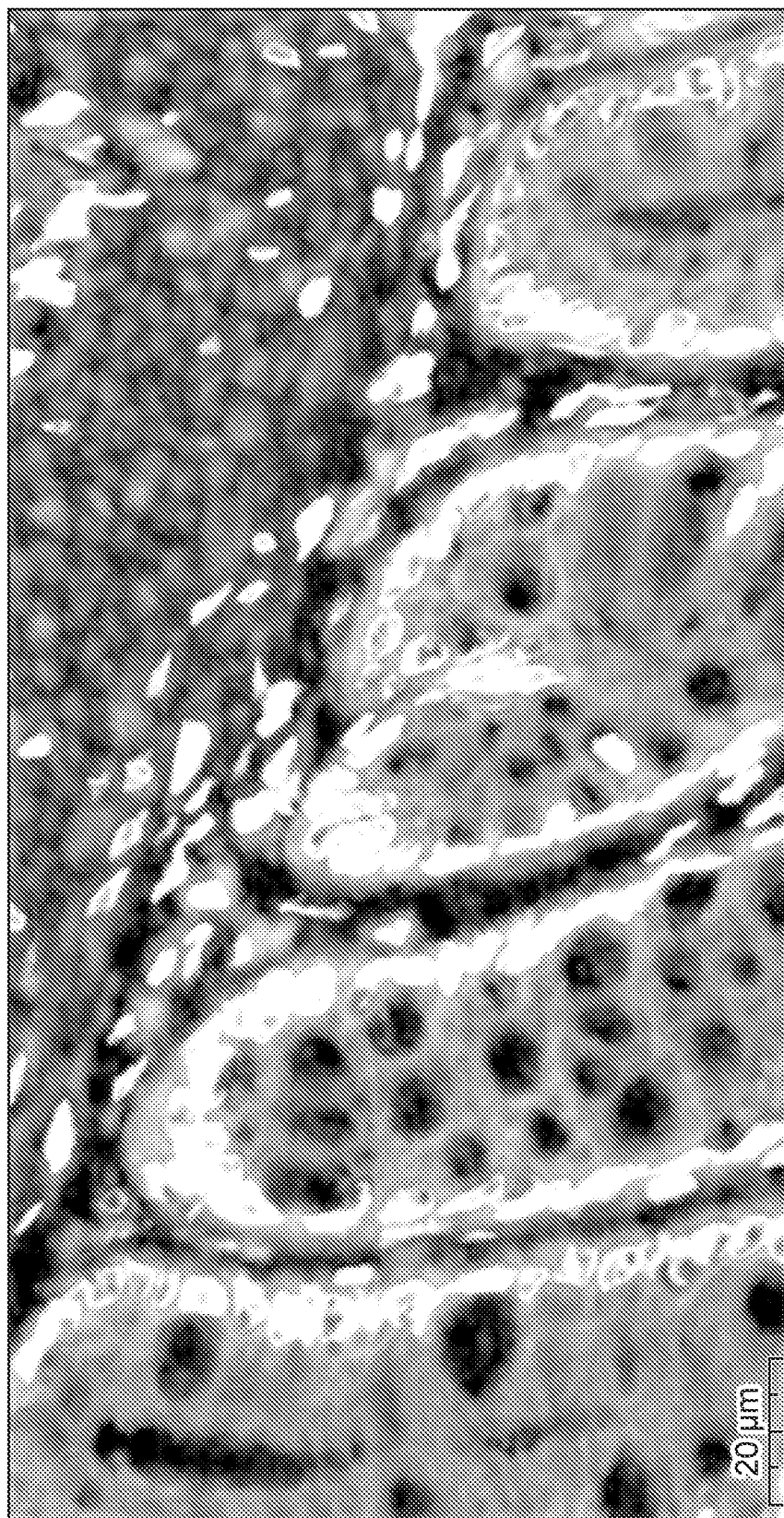


FIG. 4

# INTERNATIONAL SEARCH REPORT

International application No  
**PCT/US2022/012307**

## A. CLASSIFICATION OF SUBJECT MATTER

**INV. A61K31/485 A61P1/04 A61P1/06 A61P1/12 A61P25/04**  
**ADD.**

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

**A61K A61P**

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

**EPO-Internal, WPI Data, BIOSIS, EMBASE, FSTA**

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

| Category* | Citation of document, with indication, where appropriate, of the relevant passages                                                                                         | Relevant to claim No.             |
|-----------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------------------|
| <b>X</b>  | <b>WO 2010/006299 A1 (UNIV MINNESOTA [US];<br/>PORTOGHESE PHILIP S [US])<br/>14 January 2010 (2010-01-14)<br/>cited in the application<br/>the whole document</b><br>----- | <b>1-4, 9,<br/>12, 13, 15</b>     |
| <b>X</b>  | <b>WO 2019/071264 A1 (BLUE THERAPEUTICS INC<br/>[US]) 11 April 2019 (2019-04-11)<br/>the whole document</b><br>-----                                                       | <b>1-4, 9,<br/>12-15<br/>1-15</b> |
| <b>Y</b>  | <b>-----<br/>-/--</b>                                                                                                                                                      |                                   |



Further documents are listed in the continuation of Box C.



See patent family annex.

\* Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier application or patent but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art

"&" document member of the same patent family

Date of the actual completion of the international search

**15 April 2022**

Date of mailing of the international search report

**26/04/2022**

Name and mailing address of the ISA/

European Patent Office, P.B. 5818 Patentlaan 2  
NL - 2280 HV Rijswijk  
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Fax: (+31-70) 340-3016

Authorized officer

**Houyvet-Landriscina**

# INTERNATIONAL SEARCH REPORT

International application No  
PCT/US2022/012307

| C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                           |                       |
|------------------------------------------------------|-----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| Category*                                            | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                                                                                                                                                                                                                                                                        | Relevant to claim No. |
| Y                                                    | <p>A. S. YEKKIRALA ET AL:<br/> "N-naphthoyl-<math>\beta</math>-naltrexamine (NNTA), a highly selective and potent activator of mu/k-opioid heteromers",<br/> PROCEEDINGS OF THE NATIONAL ACADEMY OF SCIENCES,<br/> vol. 108, no. 12,<br/> 8 March 2011 (2011-03-08), pages<br/> 5098-5103, XP055589337,<br/> ISSN: 0027-8424, DOI:<br/> 10.1073/pnas.1016277108<br/> the whole document</p> <p>-----</p>                                                                                                  | 1-15                  |
| Y                                                    | <p>US 2008/242720 A1 (MANGEL ALLEN [US])<br/> 2 October 2008 (2008-10-02)<br/> the whole document<br/> paragraph [0054]</p> <p>-----</p>                                                                                                                                                                                                                                                                                                                                                                  | 1-15                  |
| Y                                                    | <p>SENGUPTA J ET AL: "Effects of kappa opioids in the inflamed rat colon",<br/> PAIN, ELSEVIER SCIENCE PUBLISHERS,<br/> AMSTERDAM, NL,<br/> vol. 79, no. 2-3,<br/> 1 February 1999 (1999-02-01), pages<br/> 175-185, XP027336149,<br/> ISSN: 0304-3959<br/> [retrieved on 1999-02-01]<br/> the whole document</p> <p>-----</p>                                                                                                                                                                            | 1-15                  |
| Y                                                    | <p>Liu Bao-Hua ET AL: "Effects of mu and kappa opioid receptor agonists and antagonists on contraction of isolated colon strips of rats with cathartic colon INTRODUCTION",<br/> China World J Gastroenterol,<br/> 1 June 2004 (2004-06-01), pages 1672-1674,<br/> XP055912866,<br/> Retrieved from the Internet:<br/> URL:https://www.ncbi.nlm.nih.gov/pmc/articles/PMC4572778/pdf/WJG-10-1672.pdf<br/> [retrieved on 2022-04-14]<br/> abstract</p> <p>-----</p> <p style="text-align: center;">-/--</p> | 1-15                  |

# INTERNATIONAL SEARCH REPORT

International application No  
PCT/US2022/012307

| C(Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT |                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        |                       |
|------------------------------------------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|-----------------------|
| Category*                                            | Citation of document, with indication, where appropriate, of the relevant passages                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                     | Relevant to claim No. |
| Y                                                    | <p><b>SALAGA M ET AL: "Systemic Administration of Sialorphan Attenuates Experimental Colitis in Mice via Interaction With Mu and Kappa Opioid Receptors", JOURNAL OF CROHN'S AND COLITIS, vol. 11, no. 8, 1 August 2017 (2017-08-01), pages 988-998, XP055912868, NL</b></p> <p><b>ISSN: 1873-9946, DOI: 10.1093/ecco-jcc/jjx043</b></p> <p><b>Retrieved from the Internet:</b></p> <p><b>URL:https://watermark.silverchair.com/jjx043.pdf?token=AQECAHi208BE49Ooan9kKhW_Ercy7Dm3ZL_9Cf3qfKAc485ysgAAAuEwggLdBgkqhkiG9w0BBwagggLOMIICygIBADCCAsMGCSqGSib3DQEHATAeBglghkgBZQMEAS4wEQQMeWCKiNgvZhodLGkAgEQgIIClHwSkMEJlYC3ZVtaSiM3RPAgyK-e8aKbocbOZ37dc oqiK1DfaOp_IJa6ccTStkYseXJXSVfUqeWEuE852ZF177tqfQq98&gt;</b></p> <p><b>abstract</b></p> <p style="text-align: center;">-----</p> | 1-15                  |

# INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No

PCT/US2022/012307

| Patent document<br>cited in search report | Publication<br>date | Patent family<br>member(s)                                                                                                                                                                                                                                                                                                                                                                                                                                         | Publication<br>date                                                                                                                                                                                                                                                                                                                                        |
|-------------------------------------------|---------------------|--------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|
| WO 2010006299 A1                          | 14-01-2010          | US 2011251227 A1<br>WO 2010006299 A1                                                                                                                                                                                                                                                                                                                                                                                                                               | 13-10-2011<br>14-01-2010                                                                                                                                                                                                                                                                                                                                   |
| WO 2019071264 A1                          | 11-04-2019          | AU 2018345909 A1<br>CA 3076233 A1<br>CN 111757738 A<br>EP 3691644 A1<br>JP 2020536891 A<br>US 2020289500 A1<br>WO 2019071264 A1                                                                                                                                                                                                                                                                                                                                    | 09-04-2020<br>11-04-2019<br>09-10-2020<br>12-08-2020<br>17-12-2020<br>17-09-2020<br>11-04-2019                                                                                                                                                                                                                                                             |
| US 2008242720 A1                          | 02-10-2008          | AU 2008232954 A1<br>BR PI0809651 A2<br>CA 2682608 A1<br>CN 101677997 A<br>CN 102641265 A<br>CY 1113317 T1<br>DK 2136801 T3<br>EP 2136801 A1<br>EP 2561870 A1<br>EP 2636407 A2<br>ES 2393815 T3<br>HK 1139860 A1<br>HR P20120798 T1<br>JP 5416085 B2<br>JP 2010523492 A<br>JP 2013173790 A<br>KR 20100016004 A<br>KR 20120058614 A<br>PL 2136801 T3<br>PT 2136801 E<br>RU 2009140042 A<br>SI 2136801 T1<br>US 2008242720 A1<br>US 2011046174 A1<br>WO 2008121496 A1 | 09-10-2008<br>07-10-2014<br>09-10-2008<br>24-03-2010<br>22-08-2012<br>13-04-2016<br>17-12-2012<br>30-12-2009<br>27-02-2013<br>11-09-2013<br>28-12-2012<br>30-09-2010<br>30-11-2012<br>12-02-2014<br>15-07-2010<br>05-09-2013<br>12-02-2010<br>07-06-2012<br>31-01-2013<br>06-12-2012<br>10-05-2011<br>31-01-2013<br>02-10-2008<br>24-02-2011<br>09-10-2008 |