

IONIC LIQUID FORMULATIONS FOR TREATING DIABETES

RELATED APPLICATIONS

[0001] This application claims the benefit of U.S. Provisional Patent Application No. 63/073,172 filed on September 01, 2020, U.S. Provisional Patent Application No. 63/154,461 filed on February 26, 2021, and U.S. Provisional Patent Application No. 63/160,575 filed on March 12, 2021, the entire contents of each of which are hereby incorporated by reference.

BACKGROUND OF THE INVENTION

[0002] Diabetes is a metabolic disease characterized by the inability of the pancreas to secrete a level of insulin adequate to maintain a normal level of systemic glucose. Despite advances, there remains a need for novel treatments of diabetes.

SUMMARY OF THE INVENTION

[0003] Provided herein, in one aspect, is a method of treating a disease or disorder in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of a composition comprising an ionic liquid.

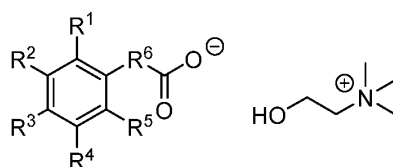
[0004] In some embodiments, the disease or disorder is diabetes. In some embodiments, the disease or disorder is Type 1 diabetes. In some embodiments, the disease or disorder is Type 2 diabetes. In some embodiments, the disease or disorder is non-alcoholic steatohepatitis.

[0005] Provided herein, in another aspect, is a method of treating obesity, preventing weight gain, or reducing weight in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of a composition comprising an ionic liquid.

[0006] In some embodiments, the composition is administered via subcutaneous, intravenous, or oral administration. In some embodiments, the composition is administered via oral administration. In some embodiments, the composition is administered as a liquid-filled capsule. In some embodiments, the composition is administered in a single dose. In some embodiments, the composition is administered in multiple doses. In some embodiments, the composition is administered to a mucus membrane.

[0007] In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.1% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.05M. In some embodiments, the ionic liquid comprises a cation:anion ratio of from about 4:1 to about 1:4.

[0008] In some embodiments, the ionic liquid is represented by Formula (I):



Formula (I),

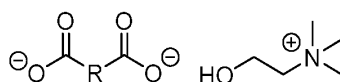
wherein:

R^1 , R^2 , R^3 , R^4 , and R^5 are independently selected from hydrogen, halo, cyano, nitro, amino, C_{1-6} alkoxy, C_{1-6} heteroalkyl, C_{1-6} haloalkyl, C_{1-6} alkyl, C_{2-6} alkenyl, and C_{2-6} alkynyl; and R^6 is selected from C_{1-6} alkyl, C_{2-6} alkenyl, and C_{2-6} alkynyl.

[0009] In some embodiments, at least two of R^1 , R^2 , R^3 , R^4 , and R^5 are hydrogen. In some embodiments, at least three of R^1 , R^2 , R^3 , R^4 , and R^5 are hydrogen. In some embodiments, R^1 , R^2 , R^3 , R^4 , and R^5 are hydrogen.

[0010] In some embodiments, R^6 is selected from C_{1-6} alkyl and C_{2-6} alkenyl. In some embodiments, R^6 is C_{1-6} alkyl. In some embodiments, R^6 is C_2 alkyl. In some embodiments, R^6 is C_{1-6} alkenyl. In some embodiments, R^6 is C_2 alkenyl.

[0011] In some embodiments, the ionic liquid is represented by Formula (II):



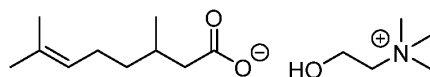
Formula (II),

wherein:

R is selected from C_{1-6} alkyl, C_{2-6} alkenyl, and C_{2-6} alkynyl.

[0012] In some embodiments, R is C_{1-6} alkyl. In some embodiments, R is C_1 alkyl. In some embodiments, R is C_3 alkyl.

[0013] In some embodiments, the ionic liquid is represented by Formula (III):



Formula (III).

[0014] In some embodiments, the ionic liquid comprises a cholinium cation and an anion selected from cinnamate, hydrocinnamate, malonate, citronellate, and glutarate. In some embodiments, the anion is selected from cinnamate, hydrocinnamate, and citronellate.

[0015] In some embodiments, the composition further comprises one or more additional agents. In some embodiments, the one or more additional agents are selected from a nucleic acid, a small molecule, and a polypeptide. In some embodiments, the one or more additional agents comprise a

nucleic acid. In some embodiments, the one or more additional agents comprise a small molecule. In some embodiments, the one or more additional agents comprise a polypeptide.

[0016] In some embodiments, the one or more additional agents are selected from a glucagon-like peptide (GLP-1), a glucagon-like peptide derivative, and a glucagon-like peptide mimetic. In some embodiments, the one or more additional agents are selected from liraglutide, exenatide, and semaglutide. In some embodiments, the one or more additional agents comprise liraglutide.

[0017] In some embodiments, the one or more additional agents are selected from insulin and pramlintide.

[0018] In some embodiments, the composition further comprises a pharmaceutically acceptable excipient.

[0019] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and hydrocinnamic acid in a 1:2 molar ratio.

[0020] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and cinnamic acid in a 1:1 molar ratio.

[0021] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and glutaric acid in a 1:1 molar ratio.

[0022] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and malonic acid in a 1:1 molar ratio.

[0023] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and octenoic acid in a 1:1 molar ratio.

[0024] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and octenoic acid in a 1:2 molar ratio.

[0025] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and citronellic acid in a 1:1 molar ratio.

[0026] Provided herein, in another aspect, is a composition comprising an ionic liquid, wherein the ionic liquid comprises choline and citronellic acid in a 1:2 molar ratio.

[0027] In some embodiments, the composition further comprises one or more additional agents. In some embodiments, the one or more additional agents are selected from a nucleic acid, a small molecule, and a polypeptide. In some embodiments, the one or more additional agents comprise a nucleic acid. In some embodiments, the one or more additional agents comprise a small molecule. In some embodiments, the one or more additional agents comprise a polypeptide.

[0028] In some embodiments, the one or more additional agents are selected from a glucagon-like peptide (GLP-1), a glucagon-like peptide derivative, and a glucagon-like peptide mimetic. In some

embodiments, the one or more additional agents are selected from liraglutide, exenatide, and semaglutide. In some embodiments, the one or more additional agents comprise liraglutide.

[0029] In some embodiments, the one or more additional agents are selected from insulin and pramlintide.

[0030] Provided herein, in another aspect, is a pharmaceutical composition comprising a composition described herein and a pharmaceutically acceptable excipient

INCORPORATION BY REFERENCE

[0031] All publications, patents, and patent applications mentioned in this specification are herein incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference.

BRIEF DESCRIPTION OF THE DRAWINGS

[0032] The novel features of the invention are set forth with particularity in the appended claims. A better understanding of the features and advantages of the present invention will be obtained by reference to the following detailed description that sets forth illustrative embodiments, in which the principles of the invention are utilized, and the accompanying drawings of which:

[0033] **FIG. 1** shows the amino acid sequence of glucagon-like peptide-1 (GLP-1).

[0034] **FIG. 2** shows the amino acid sequence of Exenatide.

[0035] **FIG. 3** shows the amino acid sequence of Liraglutide.

[0036] **FIG. 4** shows the amino acid sequence of Semaglutide.

[0037] **FIG. 5** shows various ionic liquids which are liquid at room temperature.

[0038] **FIG. 6** shows various ionic liquids which are not liquid at room temperature.

[0039] **FIG. 7** shows the change in blood glucose levels over time following intrajeunal administration of choline-citronellic acid to non-diabetic rats.

[0040] **FIG. 8** shows the change in blood glucose levels over time following administration of choline-octanoic acid and choline-octenoic acid to non-diabetic rats.

[0041] **FIG. 9** shows the change in blood glucose levels over time following administration of citronellic acid to non-diabetic rats.

[0042] **FIG. 10** shows the change in blood glucose levels over time following subcutaneous and oral administration of choline-citronellic acid to non-diabetic rats.

[0043] **FIG. 11** shows the change in blood glucose levels over time following oral administration of choline-citronellic acid to diabetic rats.

[0044] **FIG. 12** shows the change in plasma insulin levels over time following intrajeunal, subcutaneous, and oral administration of choline-citronellic acid to diabetic rats.

[0045] FIG. 13 shows the change in blood glucose levels and glucose levels in the urine over time following intrajejunal administration of choline-citronellic acid to non-diabetic rats.

[0046] FIG. 14 shows the change in serum liraglutide levels over time following intrajejunal administration of choline-hydrocinnamic acid and liraglutide to non-diabetic rats.

[0047] FIG. 15 shows Liraglutide delivery in C-Cinnamic acid 1:1 to the duodenum (liquid) or stomach of dogs (liquid or capsule) compared to IV (intravenous), SC (subcutaneous) dosing and oral unformulated Liraglutide to the stomach.

[0048] FIG. 16 shows Exen in C-Cinnamic acid 1:1 to stomach of dogs as a liquid compared to IV, SC dosing and unformulated (Exenatide-Saline).

[0049] FIG. 17 shows Semaglutide in Choline-Cinnamic Acid 1:1 delivered to the stomach in 0, 00 or 000 gelatin capsules coated with Evonik EPO coating or 0 HPMC capsules.

[0050] FIG. 18 shows Semaglutide delivery in C-Cinnamic acid 1:1 to the stomach of dogs (liquid and capsule) compared to IV, SC dosing and oral unformulated (PPB) or with SNAC (SNAC-PPB).

[0051] FIG. 19 shows co-delivery of Liraglutide and Exenatide with Choline-Cinnamic Acid 1:1.

[0052] FIG. 20 shows H&E-stained GI (gastrointestinal) tract tissues for Ionic Liquid (IL) and saline dosed rats at 100 μ L.

[0053] FIG. 21 shows blood and plasma results for 100 μ L Ionic Liquid (IL)-dosed (light grey; left columns) and saline-dosed (dark grey; right columns) rats.

[0054] FIG. 22 shows immunohistochemistry staining of jejunum tight junctions from Ionic Liquid (IL)-dose and saline-dosed (placebo) groups stained for Occuldin and Claudin-1.

[0055] FIG. 23 shows body weights of rats in 100 μ L dosed Ionic Liquid (IL) and placebo group.

DETAILED DESCRIPTION OF THE INVENTION

Definitions

[0056] Unless defined otherwise, all technical and scientific terms used herein have the same meaning as is commonly understood by one of skill in the art to which this disclosure belongs.

[0057] As used herein, the singular form “a”, “an” and “the” includes plural references unless the context clearly dictates otherwise.

[0058] The term “C_{x-y}” when used in conjunction with a chemical moiety, such as alkyl, alkenyl, or alkynyl is meant to include groups that contain from x to y carbons in the chain. For example, the term “C₁₋₆alkyl” refers to substituted or unsubstituted saturated hydrocarbon groups, including straight-chain alkyl and branched-chain alkyl groups that contain from 1 to 6 carbons. The term –C_{x-y}alkylene– refers to a substituted or unsubstituted alkylene chain with from x to y carbons in the alkylene chain. For example –C₁₋₆alkylene– may be selected from methylene, ethylene, propylene, butylene, pentylene, and hexylene, any one of which is optionally substituted.

[0059] “Alkyl” refers to substituted or unsubstituted saturated hydrocarbon groups, including straight-chain alkyl and branched-chain alkyl groups. An alkyl group may contain from one to twelve carbon atoms (*e.g.*, C₁₋₁₂ alkyl), such as one to eight carbon atoms (C₁₋₈ alkyl) or one to six carbon atoms (C₁₋₆ alkyl). Exemplary alkyl groups include methyl, ethyl, n-propyl, isopropyl, n-butyl, isobutyl, sec-butyl, *tert*-butyl, pentyl, isopentyl, neopentyl, hexyl, heptyl, octyl, nonyl, and decyl. An alkyl group is attached to the rest of the molecule by a single bond. Unless stated otherwise specifically in the specification, an alkyl group is optionally substituted by one or more substituents such as those substituents described herein.

[0060] “Haloalkyl” refers to an alkyl group that is substituted by one or more halogens. Exemplary haloalkyl groups include trifluoromethyl, difluoromethyl, trichloromethyl, 2,2,2-trifluoroethyl, 1,2-difluoroethyl, 3-bromo-2-fluoropropyl, and 1,2-dibromoethyl.

[0061] “Alkenyl” refers to substituted or unsubstituted hydrocarbon groups, including straight-chain or branched-chain alkenyl groups containing at least one double bond. An alkenyl group may contain from two to twelve carbon atoms (*e.g.*, C₂₋₁₂ alkenyl). Exemplary alkenyl groups include ethenyl (*i.e.*, vinyl), prop-1-enyl, but-1-enyl, pent-1-enyl, penta-1,4-dienyl, and the like. Unless stated otherwise specifically in the specification, an alkenyl group is optionally substituted by one or more substituents such as those substituents described herein.

[0062] “Alkynyl” refers to substituted or unsubstituted hydrocarbon groups, including straight-chain or branched-chain alkynyl groups containing at least one triple bond. An alkynyl group may contain from two to twelve carbon atoms (*e.g.*, C₂₋₁₂ alkynyl). Exemplary alkynyl groups include ethynyl, propynyl, butynyl, pentynyl, hexynyl, and the like. Unless stated otherwise specifically in the specification, an alkynyl group is optionally substituted by one or more substituents such as those substituents described herein.

[0063] “Heteroalkyl”, “heteroalkenyl” and “heteroalkynyl” refer to substituted or unsubstituted alkyl, alkenyl and alkynyl groups which respectively have one or more skeletal chain atoms selected from an atom other than carbon. Exemplary skeletal chain atoms selected from an atom other than carbon include, *e.g.*, O, N, P, Si, S, or combinations thereof, wherein the nitrogen, phosphorus, and sulfur atoms may optionally be oxidized and the nitrogen heteroatom may optionally be quaternized. If given, a numerical range refers to the chain length in total. For example, a 3- to 8-membered heteroalkyl has a chain length of 3 to 8 atoms. Connection to the rest of the molecule may be through either a heteroatom or a carbon in the heteroalkyl, heteroalkenyl or heteroalkynyl chain. Unless stated otherwise specifically in the specification, a heteroalkyl, heteroalkenyl, or heteroalkynyl group is optionally substituted by one or more substituents such as those substituents described herein.

[0064] The term "ionic liquids" as used herein refers to organic salts or mixtures of organic salts which exist in a liquid state. Ionic liquids have been shown to be useful in a variety of fields, including in industrial processing, catalysis, pharmaceuticals, and electrochemistry. The ionic liquids contain at least one anionic and at least one cationic component. Ionic liquids can comprise an additional hydrogen bond donor (*i.e.* any molecule that can provide an -OH or an -NH group); examples include but are not limited to alcohols, fatty acids, and amines. The anionic and the cationic component may be present in any molar ratio. Exemplary molar ratios (cation: anion) include but are not limited to 1:1, 1:2, 2:1, and ranges between these ratios. In some embodiments, the ionic liquid or solvent exists as a liquid below 100 °C. In some embodiments, the ionic liquid or solvent exists as a liquid at room temperature.

[0065] The phrase "pharmaceutically acceptable excipient" or "pharmaceutically acceptable carrier" as used herein means a pharmaceutically acceptable material, composition or vehicle, such as a liquid or solid filler, diluent, excipient, solvent, or encapsulating material. Each carrier must be "acceptable" in the sense of being compatible with the other ingredients of the formulation and not injurious to the patient. Some examples of materials which can serve as pharmaceutically acceptable carriers include: (1) sugars, such as lactose, glucose and sucrose; (2) starches, such as corn starch and potato starch; (3) cellulose, and its derivatives, such as sodium carboxymethyl cellulose, ethyl cellulose and cellulose acetate; (4) powdered tragacanth; (5) malt; (6) gelatin; (7) talc; (8) excipients, such as cocoa butter and suppository waxes; (9) oils, such as peanut oil, cottonseed oil, safflower oil, sesame oil, olive oil, corn oil and soybean oil; (10) glycols, such as propylene glycol; (11) polyols, such as glycerin, sorbitol, mannitol and polyethylene glycol; (12) esters, such as ethyl oleate and ethyl laurate; (13) agar; (14) buffering agents, such as magnesium hydroxide and aluminum hydroxide; (15) alginic acid; (16) pyrogen-free water; (17) isotonic saline; (18) Ringer's solution; (19) ethyl alcohol; (20) phosphate buffer solutions; and (21) other non-toxic compatible substances employed in pharmaceutical formulations.

[0066] The term "effective amount" or "therapeutically effective amount" refers to that amount of a compound described herein that is sufficient to affect the intended application, including but not limited to disease treatment, as defined below. The therapeutically effective amount may vary depending upon the intended treatment application (*in vivo*), or the subject and disease condition being treated, *e.g.*, the weight and age of the subject, the severity of the disease condition, the manner of administration and the like, which can readily be determined by one of ordinary skill in the art. The term also applies to a dose that will induce a particular response in target cells, *e.g.*, reduction of platelet adhesion and/or cell migration. The specific dose will vary depending on the particular compounds chosen, the dosing regimen to be followed, whether it is administered in

combination with other compounds, timing of administration, the tissue to which it is administered, and the physical delivery system in which it is carried.

[0067] As used herein, “treatment” or “treating” refers to an approach for obtaining beneficial or desired results with respect to a disease, disorder, or medical condition including but not limited to a therapeutic benefit and/or a prophylactic benefit. A therapeutic benefit can include, for example, the eradication or amelioration of the underlying disorder being treated. Also, a therapeutic benefit can include, for example, the eradication or amelioration of one or more of the physiological symptoms associated with the underlying disorder such that an improvement is observed in the subject, notwithstanding that the subject may still be afflicted with the underlying disorder. In certain embodiments, for prophylactic benefit, the compositions are administered to a subject at risk of developing a particular disease, or to a subject reporting one or more of the physiological symptoms of a disease, even though a diagnosis of this disease may not have been made.

[0068] A “therapeutic effect,” as that term is used herein, encompasses a therapeutic benefit and/or a prophylactic benefit as described above. A prophylactic effect includes delaying or eliminating the appearance of a disease or condition, delaying or eliminating the onset of symptoms of a disease or condition, slowing, halting, or reversing the progression of a disease or condition, or any combination thereof.

[0069] The term “co-administration,” “administered in combination with,” and their grammatical equivalents, as used herein, encompass administration of two or more agents to an animal, including humans, so that both agents and/or their metabolites are present in the subject at the same time. Co-administration includes simultaneous administration in separate compositions, administration at different times in separate compositions, or administration in a composition in which both agents are present.

[0001] As used herein, the terms “subject” and “patient” include animals (*e.g.*, vertebrates, amphibians, fish, mammals, cats, dogs, horses, pigs, cows, sheep, rodents, rabbits, squirrels, bears, and primates (*e.g.*, chimpanzees, gorillas, and humans)). The subject is preferably a mammal. The mammal can be, *e.g.*, any mammal, *e.g.*, a human, a primate, a mouse, a rat, a dog, a cat, a horse, as well as livestock or animals grown for food consumption, *e.g.*, cattle, sheep, pigs, chickens, and goats. In a preferred embodiment, the mammal is a human.

[0002] A “control” or “standard control” refers to a sample, measurement, or value that serves as a reference, usually a known reference, for comparison to a test sample, measurement, or value. For example, a test sample can be taken from a subject having a given disease (*e.g.*, diabetes) and compared with a known normal (non-diseased) individual (*e.g.*, a standard control subject). A standard control can also represent an average measurement or value gathered from a population of

similar individuals (*e.g.*, standard control subjects) that do not have a given disease (*i.e.*, standard control population), *e.g.*, healthy individuals with a similar medical background, same age, weight, etc. A standard control value can also be obtained from the same individual, *e.g.*, from an earlier-obtained sample from the patient prior to disease onset. For example, a control can be devised to compare therapeutic benefit based on pharmacological data (*e.g.*, half-life) or therapeutic measures (*e.g.*, comparison of side effects). Controls are also valuable for determining the significance of data. For example, if values for a given parameter are widely variant in controls, variation in test samples will not be considered as significant. One of skill will recognize that standard controls can be designed for assessment of any number of parameters.

[0070] As used herein, a “drug” is any agent which will exert an effect on a target cell or organism. A drug can be selected from a group comprising: chemicals; small organic or inorganic molecules; peptide; protein; or nucleic acid. Non-limiting examples of active compounds contemplated for use in the methods described herein include small molecules, polypeptides, nucleic acids, antibodies, vaccines, a GLP-1 polypeptide or mimetic/analog thereof, pramlintide, and insulin.

[0071] As used herein, “diabetes” refers to diabetes mellitus, a metabolic disease characterized by a deficiency or absence of insulin secretion by the pancreas. As used throughout, “diabetes” includes all types including Type 1 and Type 2 diabetes mellitus unless otherwise specified herein. The two most common forms of diabetes are due to either a diminished production of insulin (in Type 1), or diminished response by the body to insulin (in Type 2). In Type 1 diabetes, the function of the pancreas is progressively lost, thus eventually making the patient entirely dependent on the exogenously delivered insulin for the management of diabetes. In Type 2, the patient maintains some functioning of the pancreas, but the sensitivity of the body to insulin is reduced, thus reducing the extent of glycemia maintained by the patient. Type 2 patients are treated by a variety of drugs including oral medications that increase glucose sensitivity, GLP-1 analogs, or insulin. Both types of diabetes lead to hyperglycemia, which causes the acute signs of diabetes: excessive urine production, increased thirst and increased fluid intake, blurred vision, unexplained weight loss, lethargy, and changes in energy metabolism. Diabetes can cause many complications including neuropathy, retinopathy, poor microvascular function, renal failure, and poor wound healing. A “pre-diabetic” subject can be characterized, for example, as having elevated fasting blood sugar or elevated post-prandial blood sugar such that the glucose levels do not fit the current medical definitions of diabetes. A “newly diagnosed” subject refers to a Type 1 diabetic patient that is within 1-3 years of their diagnosis. This patient population can be physiologically or emotionally different from the general Type 1 diabetic population.

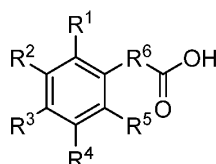
[0072] The term "obesity" refers to excess fat in the body. Obesity can be determined by any measure accepted and utilized by those of skill in the art. Currently, an accepted measure of obesity is body mass index (BMI). Consequences of obesity include cardiovascular disease, high blood pressure (*i.e.*, hypertension), osteoarthritis, cancer, and diabetes.

Compositions Comprising Ionic Liquids

[0073] Provided herein, in some embodiments, are compositions comprising ionic liquids useful in the treatment of certain diseases and disorders. In some embodiments, the anion in the ionic liquid may be chosen from cinnamic acid, hydrocinnamic acid, hydroxycinnamic (3-phenylpropanoic or benzylacetic) acid, methoxycinnamic acid, ferulic acid, isoferulic acid, 2-phenylpropionic (hydratropic acid), coumaric acid, 3,3-diphenylpropionic acid, 3,5-dimethoxy-4-hydroxy-cinnamic acid (sinapinic acid). Other structural analogs of cinnamic acid may be used.

[0074] In some embodiments, the ionic liquid comprises choline and hydrocinnamic acid in a 1:2 molar ratio. In some embodiments, the ionic liquid comprises choline and cinnamic acid in a 1:1 molar ratio. In some embodiments, the ionic liquid comprises choline and glutaric acid in a 1:1 molar ratio. In some embodiments, the ionic liquid comprises choline and malonic acid in a 1:1 molar ratio. In some embodiments, the ionic liquid comprises choline and octenoic acid in a 1:1 molar ratio. In some embodiments, the ionic liquid comprises choline and octenoic acid in a 1:2 molar ratio. In some embodiments, the ionic liquid comprises choline and citronellic acid in a 1:1 molar ratio. In some embodiments, the ionic liquid comprises choline and citronellic acid in a 1:2 molar ratio.

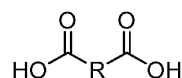
[0075] In some embodiments, a structural analog of cinnamic acid is represented by the formula:



wherein:

R^1 , R^2 , R^3 , R^4 , and R^5 are independently selected from hydrogen, halo, cyano, nitro, amino, C_{1-6} alkoxy, C_{1-6} heteroalkyl, C_{1-6} haloalkyl, C_{1-6} alkyl, C_{2-6} alkenyl, and C_{2-6} alkynyl; and R^6 is selected from C_{1-6} alkyl, C_{2-6} alkenyl, and C_{2-6} alkynyl.

[0076] In some embodiments, the anion is a diacid represented by the formula:

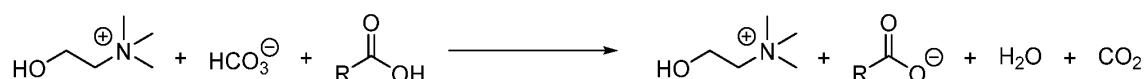


wherein:

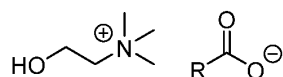
R is selected from C₁₋₆alkyl, C₂₋₆alkenyl, and C₂₋₆alkynyl.

[0077] Choline (or cholinium) is a suitable choice of cation in the preparation of ionic liquids. However, the cation can be chosen from a variety of molecules including salts of choline (e.g., choline chloride), derivatives of choline, or any other biocompatible cation that is able to form an ionic liquid to with the anions described herein.

[0078] In some embodiments, the ionic liquid is prepared by mixing an acid with choline bicarbonate, as exemplified by the scheme below:



[0079] Choline bicarbonate reacts with a carboxylic acid to form water, carbon dioxide, and an ionic liquid represented by the formula:



[0080] Depending on the ratio of anion and cation in the reaction mixture, the resultant mixture may also contain either excess acid or excess choline bicarbonate. The term “ionic liquid” used herein includes all stoichiometries, including equimolar acid and choline carbonate, excess acid, or excess choline bicarbonate.

[0081] The ionic liquid structures shown with or without an acidic proton are equivalent and interchangeable depending on the concentration and composition.

[0082] In some embodiments, the properties of an ionic liquid are determined by the ionic interactions between the anion and the cation. In some embodiments, the properties of an ionic liquid are determined by the hydrogen bonding interactions between the anion and cation. The relative contribution of ionic and hydrogen bonding interactions to the properties of the ionic liquid may vary depending on the nature of the ions.

[0083] In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.01% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.02% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.03% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.04% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.05% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.06% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.07%

concentration of at least 0.8M. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.9M. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 1.0M.

[illegible]

[illegible]

[0086] In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.1% weight per volume. In some embodiments, the composition comprises the ionic liquid at a concentration of at least 0.05 M.

[0087] In some embodiments, the ionic liquid comprises a cation:anion ratio of from about 2:1 to about 1:2. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 2.2:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 2.1:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 2:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.9:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.8:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.7:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.6:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.5:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.4:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.3:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.2:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1.1:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.2. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.3. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.4. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.5. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.6. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.7. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.8. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:1.9. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:2. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:2.1. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:2.2. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:2.3. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:2.4. In some embodiments, the ionic liquid comprises a cation:anion ratio of about 1:2.5.

[0088] In some embodiments, the ionic liquid comprises any one of the cations listed in Table 1. In some embodiments, the ionic liquid comprises any one or more of the cations listed in Table 1. In some embodiments, the ionic liquid comprises any one of the anions listed in Table 1. In some embodiments, the ionic liquid comprises any one or more of the anions listed in Table 1. In some embodiments, the ionic liquid comprises any one of the cation:anion ratio listed in Table 1. In some embodiments, the composition as provided herein comprises any one of the ionic liquids listed in Table 1. In some embodiments, the composition as provided herein comprises any one or more of the ionic liquids listed in Table 1. In some embodiments, the composition as provided herein

comprises any one of the ionic liquids in any one of the cation:anion ratios listed in Table 1. In some embodiments, the composition as provided herein comprises any one or more of the ionic liquids in any one of the cation:anion ratios listed in Table 1.

Formulation

[0089] In some embodiments, an ionic liquid provided herein is formulated in combination with a one or more drugs. In some embodiments, the ionic liquid can be combined with another solvent to enhance solubility and/or delivery. The solvent may be aqueous or non-aqueous. In some embodiments, the purpose of the solvent is to control the dose of the ionic liquid experienced by the mucus membrane or the gastrointestinal tract. Dilution of the ionic liquid by the solvent can serve the purpose of delivering a safe dose to the subject. In some embodiments, the purpose of the solvent is to improve solubility of the one or more drugs. Such improvements may come from the ability of the solvent to control the physicochemical environment of the ionic liquid to match the chemical properties of the one or more drugs. In some embodiments, the solvent may serve the purpose of improving the delivery across the mucosal membrane.

[0090] The solvents used may include without limitation: sterile water, saline solution, glycerin, propylene glycol, ethanol, oils, ethyl oleate, isopropyl myristate, benzyl benzoate, or surfactants.

[0091] In some embodiments, the solvent is chosen so as to not adversely impact the compatibility of the ionic liquid with the capsule.

[0092] In some embodiments, the one or more drugs may form micelles or other self-assembled structures. In some embodiments, such structures may occur only in the presence of ionic liquids.

[0093] In some embodiments, the one or more drugs is a nucleic acid molecule. A nucleic acid molecule, as described herein, can be a vector, an expression vector, an inhibitory nucleic acid, an aptamer, a template molecule or cassette (*e.g.*, for gene editing), or a targeting molecule (*e.g.*, for CRISPR-Cas technologies), or any other natural or synthetic nucleic acid molecule intended for delivery to an organism.

[0094] In any of the embodiments, the one or more drugs may be designed with the intent of treating a local tissue, *e.g.*, the mucosal membrane of the intestine, treating a distant tissue, *e.g.*, the liver, or entering systemic circulation.

[0095] In some embodiments, a composition as described herein, *e.g.*, a composition comprising ionic liquids and one or more drugs, can further comprise a pharmaceutically acceptable excipient. Suitable excipients include, for example, water, saline, glycerol, ethanol, or the like, and combinations thereof. In addition, if desired, the composition can contain minor amounts of

additional excipients such as emulsifying agents, surfactants, pH buffering agents, and the like, which enhance the effectiveness of the one or more drugs.

[0096] In some embodiments, the composition comprising an ionic liquid may be further encapsulated in a dosage form designed to facilitate delivery to an organism. Non-limiting examples of such dosage forms include capsules, tablets, and syrups.

[0097] In some embodiments, formulation may require excipients sugars (such as lactose), starches (such as corn starch), cellulose, cellulose derivatives (such as sodium carboxymethyl cellulose), gelatin, and other compatible substances.

[0098] In some embodiments, a composition comprising an ionic liquid described herein further comprises one or more additional agents. In some embodiments, the one or more additional agents are selected from a nucleic acid, a small molecule, and a polypeptide. In some embodiments, the one or more additional agents comprise a nucleic acid. In some embodiments, the one or more additional agents comprise a small molecule. In some embodiments, the one or more additional agents comprise a polypeptide. In some embodiments the polypeptide comprises an Antibody. In some embodiments, the Antibody comprises any one selected from Fragment Antigen-binding (Fab, F(ab')₂), single chain variable fragment (scFv), and Nanobodies.

[0099] In some embodiments, the one or more additional agents are selected from a glucagon-like peptide (GLP-1), a glucagon-like peptide derivative, and a glucagon-like peptide mimetic. In some embodiments, the one or more additional agents are selected from liraglutide, exenatide, and semaglutide. In some embodiments, the one or more additional agents comprise liraglutide.

[0100] In some embodiments, the one or more additional agents are selected from insulin and pramlintide.

Ionic Liquids for the Treatment of Diseases and Disorders

[0101] Provided herein, in some embodiments, is a method of treating a metabolic disease or disorder in a subject in need thereof, comprising administering a composition comprising an ionic liquid. Metabolic disorders include but are not limited to obesity, diabetes, fatty liver disease, or non-alcoholic fatty liver disease.

[0102] Provided herein, in some embodiments, is the use of ionic liquids for treating diabetes by oral administration. Oral administration can be achieved in any one of the dosing forms including pills, caplets, capsules, aerosol sprays, or liquids. The ionic liquid or the one or more drugs to be delivered with the ionic liquid can be encapsulated in a capsule. The ionic liquid with the dosing form may be present in any of the physical forms including a clear neat ionic liquid, a homogenous mixture of an ionic liquid with a pharmaceutically acceptable diluent, an emulsion, or a suspension.

The oral dose can also be given as a syrup, a spray, or an aerosol. The composition of any oral dose disclosed herein may contain a predetermined amount of ionic liquid and optionally one or more drugs, and may be prepared by methods of pharmacy well known to those skilled in the art.

[0103] In some embodiments, described herein is a method of treatment of diabetes comprising orally administering an oral formulation of insulin in combination with an ionic liquid.

[0104] In some embodiments, described herein is a method of treatment of diabetes comprising orally administering an oral formulation of insulin and pramlintide in combination with an ionic liquid.

[0105] In some embodiments, described herein is a method of treatment of diabetes comprising orally administering an oral formulation of liraglutide or exenatide in an ionic liquid.

[0106] As described herein, ionic liquids are able to safely carry active compounds across the mucus membranes encountered during oral administration.

[0107] As described in the examples herein, when administered together with one or more drugs, ionic liquids solubilize the one or more drugs and result in enhanced delivery into systemic circulation. Accordingly, they are particularly suitable as delivery vehicles to and/or across mucus membranes.

[0108] In some embodiments, provided herein is a method of delivery of one or more drugs, the method comprising administering the one or more drugs in combination with an ionic liquid to a mucus membrane, *e.g.*, a nasal, oral, or vaginal membrane.

[0109] In some embodiments, provided herein is a method of delivery of one or more drugs, the method comprising administering the one or more drugs at the dose of at least 0.01 mg/kg, 0.02 mg/kg, 0.03 mg/kg, 0.04 mg/kg, 0.05 mg/kg, 0.06 mg/kg, 0.07 mg/kg, 0.08 mg/kg, 0.09 mg/kg, 0.1 mg/kg, 0.2 mg/kg, 0.3 mg/kg, 0.4 mg/kg, 0.5 mg/kg, 0.6 mg/kg, 0.7 mg/kg, 0.8 mg/kg, 0.9 mg/kg, 1.0 mg/kg, 2.0 mg/kg, 3.0 mg/kg, 4.0 mg/kg, 5.0 mg/kg, 6.0 mg/kg, 7.0 mg/kg, 8.0 mg/kg, 9.0 mg/kg, 10 mg/kg, 20 mg/kg, 30 mg/kg, 40 mg/kg, 50 mg/kg, 60 mg/kg, 70 mg/kg, 80 mg/kg, 90 mg/kg, 100 mg/kg, 150 mg/kg, 200 mg/kg, 250 mg/kg, 300 mg/kg, 350 mg/kg, 400 mg/kg, 450 mg/kg, 500 mg/kg, 550 mg/kg, 600 mg/kg, 650 mg/kg, 700 mg/kg, 750 mg/kg, 800 mg/kg, 850 mg/kg, 900 mg/kg, 950 mg/kg, 1000 mg/kg, 1100 mg/kg, 1200 mg/kg, 1300 mg/kg, 1400 mg/kg, 1500 mg/kg, 1600 mg/kg, 1700 mg/kg, 1800 mg/kg, 1900 mg/kg, 2000 mg/kg, 2500 mg/kg, 3000 mg/kg, 3500 mg/kg, 4000 mg/kg, 4500 mg/kg, 5000 mg/kg, 5500 mg/kg, 6000 mg/kg, 6500 mg/kg, 7000 mg/kg, 7500 mg/kg, 8000 mg/kg, 8500 mg/kg, 9000 mg/kg, or 10000 mg/kg. In some embodiments, provided herein is a method of delivery of one or more drugs, the method comprising administering the one or more drugs at the dose of 0.01 mg/kg, 0.02 mg/kg, 0.03 mg/kg, 0.04 mg/kg, 0.05 mg/kg, 0.06 mg/kg, 0.07 mg/kg, 0.08 mg/kg, 0.09 mg/kg, 0.1 mg/kg, 0.2

mg/kg, 0.3 mg/kg, 0.4 mg/kg, 0.5 mg/kg, 0.6 mg/kg, 0.7 mg/kg, 0.8 mg/kg, 0.9 mg/kg, 1.0 mg/kg, 2.0 mg/kg, 3.0 mg/kg, 4.0 mg/kg, 5.0 mg/kg, 6.0 mg/kg, 7.0 mg/kg, 8.0 mg/kg, 9.0 mg/kg, 10 mg/kg, 20 mg/kg, 30 mg/kg, 40 mg/kg, 50 mg/kg, 60 mg/kg, 70 mg/kg, 80 mg/kg, 90 mg/kg, 100 mg/kg, 150 mg/kg, 200 mg/kg, 250 mg/kg, 300 mg/kg, 350 mg/kg, 400 mg/kg, 450 mg/kg, 500 mg/kg, 550 mg/kg, 600 mg/kg, 650 mg/kg, 700 mg/kg, 750 mg/kg, 800 mg/kg, 850 mg/kg, 900 mg/kg, 950 mg/kg, 1000 mg/kg, 1100 mg/kg, 1200 mg/kg, 1300 mg/kg, 1400 mg/kg, 1500 mg/kg, 1600 mg/kg, 1700 mg/kg, 1800 mg/kg, 1900 mg/kg, 2000 mg/kg, 2500 mg/kg, 3000 mg/kg, 3500 mg/kg, 4000 mg/kg, 4500 mg/kg, 5000 mg/kg, 5500 mg/kg, 6000 mg/kg, 6500 mg/kg, 7000 mg/kg, 7500 mg/kg, 8000 mg/kg, 8500 mg/kg, 9000 mg/kg, or 10000 mg/kg.

Glucagon-Like Peptide-1 (GLP-1)

[0110] Glucagon-Like Peptide-1 (GLP-1) is a peptide hormone known to reduce food intake and hunger in humans. The amino acid sequence of GLP-1 is shown in **FIG. 1**. GLP-1 is an incretin derived from the transcription product of the proglucagon gene that contributes to glucose homeostasis. Because natural GLP-1 has an extremely short half-life, making its use as a therapeutic challenging, modified versions of GLP-1 with greater stability have been developed. Such modifications can be performed either by varying the sequence of the peptide or by conjugating another entity to the peptide. A common modification includes attachment of a lipid tail. GLP-1 mimetics are currently being used in the treatment of Type 2 diabetes, with recent clinical trials demonstrating that these treatments improve glucose homeostasis. They also help in achieving weight loss.

[0111] Various GLP-1 mimetics are known and used in the treatment of diabetes. GLP-1 mimetics (or analogs) can include exenatide. The amino acid sequence of Exenatide is shown in **FIG. 2**. Other examples of GLP-1 analogs include derivatives for reducing enzymatic degradation, *e.g.*, lixisenatide, dulaglutide, semaglutide, albiglutide, liraglutide, and taspoglutide. The amino acid sequence of Liraglutide is shown in **FIG. 3**. The amino acid sequence of Semaglutide is shown in **FIG. 4**.

[0112] In some embodiments, described herein is a method of treatment of diabetes comprising orally administering an oral formulation of a GLP-1 polypeptide or mimetic/analog thereof in combination with ionic liquid.

[0113] In some embodiments, the compositions provided herein can be used to treat obesity by delivering a composition comprising an ionic liquid and a GLP-1 analog.

[0114] In some embodiments, the compositions provided herein can be used to treat obesity by dual action of the ionic liquid and the GLP-1 analog. Certain ionic liquids reduce fat absorption

across the intestinal mucosa. The net result of the composition on reduced body weight may arise from a combination of reduced fat absorption, reduced food uptake, and increased delivery of a GLP-1 analog.

EXAMPLES

General

[0115] All animal experiments were performed in accordance with the animal care committee guidelines and the Guide for the Care and Use of Animals of the Institute of Laboratory Animal Resources, National Research Council.

Example 1: Preparation of Choline Citronellate 1:2

[0116] To two equivalents of neat citronellic acid (20 g, 0.117 mol, 2 equiv.) in a 500 mL round bottom flask was added 12.13 g of an 80 wt% solution of choline bicarbonate (9.70 g, 0.059 mol, 1 equiv.). The mixture was stirred at 40 °C until CO₂ evolution ceased. Solvent was removed by rotary evaporation at 60°C for 1 hour, and the product was dried in a vacuum oven for 48 hours at 60° C.

Example 2: Preparation of Choline Cinnamate 1:2

[0117] To two equivalents of neat cinnamic acid (30 g, 0.202 mol, 2 equiv.) in a 500 mL round bottom flask was added 20.91 g of an 80 wt% solution of choline bicarbonate (16.72 g, 0.101 mol, 1 equiv.). Ethanol (5 mL) was added to mixture in order to solubilize the cinnamic acid. The mixture was stirred at 40 °C until CO₂ evolution ceased. Solvent was removed by rotary evaporation at 60°C for 1 hour, and the product was dried in a vacuum oven for 48 hours at 60° C.

Example 3: Preparation of Ionic Liquids

[0118] Several ionic liquids comprising choline as a cation and various anions were synthesized. To prepare ionic liquids, 2, 1, 0.5, or 0.33 equivalents of choline bicarbonate (80 wt% solution) were added to neat carboxylic acid anion in a 250-mL round bottom flask. For anions not miscible with the aqueous choline bicarbonate solution, a co-solvent, such as ethanol, was added until a homogenous mixture formed. The mixture was stirred at room temperature until CO₂ evolution ceased. Solvent was removed by rotary evaporation at 60 °C for 20 minutes, and each product was dried in a vacuum oven for 48 hours at 60 °C.

[0119] Using 62 different anions, 108 different ionic liquids were synthesized. Of these 108 ionic liquids, 43 were solids at room temperature and 65 were liquids at room temperature. In some instances, ionic liquids that were non-flowable waxy solids or spreadable greasy solids liquified at

elevated temperature ($>30^{\circ}\text{C}$). In some instances, solid powders did not liquefy at elevated temperature ($>30^{\circ}\text{C}$). In some instances (for example, decanoic acid), a single anion resulted in a liquid at one ratio (2:1) and in a solid at another ratio (1:1 or 1:2). The various ionic liquids formed and their physical characteristics are summarized in **Table 1**:

Table 1: Physical Properties of Ionic Liquids

Anion	Cation	Physical Form (Ratios Listed as Cation:Anion)
(R)- α -Lipoic Acid	Choline	Solid at 2:1, 1:1, 1:2,
2-(4-Isobutylphenyl)propionic Acid	Choline	Solid at 1:1
2-(4,4-Dimethyl-2-pentanyl)-5,7,7-trimethyloctanoic Acid	Choline	Solid at 1:1
2-Hexyldecanoic Acid	Choline	Liquid at 1:1
2-Hydroxyhippuric Acid	Choline	Solid at 1:1
3,7-Dimethyloctanoic Acid	Choline	Solid at 1:1, 1:2
4-Methylhexanoic Acid	Choline	Liquid at 1:1, 1:2
4-Methyloctanoic Acid	Choline	Liquid at 1:1, Solid at 1:2
4-Methylvaleric Acid	Choline	Liquid at 1:2
5-Norbornene-2-carboxylic Acid	Choline	Solid at 1:1
Abietic Acid	Choline	Solid at 1:1
Acetic Acid	Choline	Liquid at 2:1, 1:1, 1:2
Arachidonic Acid	Choline	Solid at 1:1
Caffeic Acid	Choline	Solid at 1:1
Cinnamic Acid	Choline	Liquid at 1:1, 1:1.5, 1:2
Citric Acid	Choline	Liquid at 2:1, 3:1, 4:1; Solid at 1:1
Citronellic Acid	Choline	Liquid at 2:1, 1:1, 1:2
Crotonic Acid	Choline	Liquid at 1:1, 1:2
D-(+)-Galactonic Acid	Choline	Solid at 1:1
Decanoic Acid	Choline	Liquid at 2:1; Solid at 1:1, 1:2
Deoxycholic Acid	Choline	Solid at 2:1, 1:1
Dihydrobenzoic Acid	Choline	Solid at 1:1
Eicosapentanoic Acid (EPA)	Choline	Solid at 1:1
Ellagic Acid	Choline	Solid at 1:1
Fumaric Acid	Choline	Liquid at 2:1, Solid at 1:1
Geranic Acid	Choline	Liquid at 1:1, 1:2
Glutaric Acid	Choline	Liquid at 2:1, 1:1
Glycolic Acid	Choline	Liquid at 2:1, 1:1
Hexanoic Acid	Choline	Liquid at 1:1, 1:2
Hydrocinnamic Acid (3-Phenylpropionic Acid)	Choline	Liquid at 1:1, 1:2
Isovaleric Acid	Choline	Liquid at 1:2
L-(+)-Tartaric Acid	Choline	Liquid at 2:1; Solid at 1:1
L-Ascorbic Acid	Choline	Solid at 1:1
L-Glutathione reduced	Choline	Solid at 2:1
Lactic Acid	Choline	Liquid at 1:1, 1:2
Lauric Acid	Choline	Solid at 1:1, 1:2
Levulinic Acid	Choline	Liquid at 1:1, 1:2

Anion	Cation	Physical Form (Ratios Listed as Cation:Anion)
Linoleic Acid	Choline	Liquid at 1:2; Solid at 1:1
Linolenic Acid	Choline	Liquid at 1:2
Maleic Acid	Choline	Liquid at 2:1, 1:1
Malonic Acid	Choline	Liquid at 1:1
Mesaconic Acid	Choline	Solid at 1:1; Liquid at 2:1
Nonanoic Acid	Choline	Solid at 1:1, 1:2
Octanoic Acid	Choline	Solid at 1:2, Liquid at 1:1
Oleic Acid	Choline	Solid at 1:1, 1:2
p-Toluenesulfonic Acid	Choline	Solid at 1:1; Liquid at 1:2
Perillic Acid	Choline	Solid at 1:1
Phosphoric Acid	Choline	Solid at 1:1; Liquid at 2:1, 1:2
Pimelic Acid	Choline	Liquid at 2:1, 1:1
Propionic Acid	Choline	Liquid at 2:1, 1:1
Pyroglutamic Acid	Choline	Solid at 1:1; Liquid at 1:2
Pyruvic Acid	Choline	Liquid at 1:1, 1:2
Ricinoleic Acid	Choline	Liquid at 1:1, 1:2
Sorbic Acid	Choline	Liquid at 1:1, 1:2
Syringic Acid	Choline	Solid at 1:1
Trans-2-Decenoic Acid	Choline	Solid at 1:1, 1:2
Trans-2-Hexenoic Acid	Choline	Liquid at 1:1, 1:2
Trans-2-Octenoic Acid	Choline	Liquid at 1:1, 1:2
Trans-3-Octenoic Acid	Choline	Liquid at 1:1, 1:2
Trans-7-Octenoic Acid	Choline	Liquid at 1:1, 1:2
Trans-Ferulic Acid	Choline	Solid at 1:1
Undecanoic Acid	Choline	Solid at 1:1
Vanillic Acid	Choline	Solid at 1:1
α -Ketoglutaric Acid	Choline	Liquid at 2:1
Succinic Acid	Choline	Liquid at 2:1, 1:1, 1:2
Malic Acid	Choline	Liquid at 2:1, 1:1
Mandelic Acid	Choline	Liquid at 1:1, 1:2

Example 4: Physical Form of Ionic Liquids and Deep Eutectic Solvents

[0120] Ionic liquids varied significantly in their appearance and properties. Some ionic liquids, such as choline-tartaric acid (2:1) are clear liquids at room temperature. Others, such as choline-cinnamic acid are viscous yellow liquids at room temperature. Various ionic liquids which are liquid at room temperature are shown in **FIG. 5**.

[0121] Some mixtures of anion and cation are not a liquid at room temperature. For example, choline-tartaric acid (1:1) is a solid, and choline-decanoic acid (1:1) is a waxy solid. Various non-liquid compositions are shown in **FIG. 6**. Ionic liquids that exist in a liquid form at room temperature are particularly suitable for the pharmaceutical applications described herein.

Example 5: Effect on Blood Glucose Levels Following Choline-Citronellic Acid**Administration**

[0122] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed with choline-citronellic acid via intrajejunal injection. As fasting control group was dosed with a saline solution via intrajejunal injection. About 250 μ L of blood was collected at regular intervals in order to determine the blood glucose level. The obtained values, plotted as mean percent change $\pm$ standard error in blood glucose levels with respect to initial reading (n=3) versus time, are shown in **FIG. 7**.

[0123] Choline-citronellic produced an immediate and dose-dependent decrease in blood glucose levels. Control treatment with saline did not change the blood glucose level from baseline. However, when the rats were administered 10, 20, 50 and 100 μ L of choline-citronellic acid, the blood glucose level dropped in a dose-dependent manner. For a 100 μ L dose, the glucose level dropped by about 70% from baseline. This scale of reduction in blood glucose level would likely be efficacious in the treatment of diabetes.

Example 6: Effect on Blood Glucose Levels Following Choline-Octanoic Acid and Choline-Octenoic Acid Administration

[0124] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed with 100 μ L choline-octanoic acid or choline-octenoic acid via intrajejunal injection. As fasting control group was dosed with a saline solution via intrajejunal injection. About 250 μ L of blood was collected at regular intervals in order to determine the blood glucose level. The obtained values, plotted as mean percent change $\pm$ standard error in blood glucose levels with respect to initial reading (n=3) versus time, are shown in **FIG. 8**.

[0125] Surprisingly, unlike choline-citronellic acid, which induced a pronounced decrease in blood glucose levels, neither choline-octanoic acid nor choline-octenoic acid decreased blood glucose levels in rats.

Example 7: Effect on Blood Glucose Levels Following Citronellic Acid Administration

[0126] The obtained values, plotted as mean percent change $\pm$ standard error in blood glucose levels with respect to initial reading (n=4) versus time, are shown in **FIG. 9**.

[0127] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed with choline-citronellic acid or citronellic acid alone via intrajejunal injection. A 50 μ L dose of choline-citronellic acid produced an immediate decrease in blood glucose levels to about 50% of starting levels. However, when the rats were administered 38 μ L of

citronellic acid alone, the equivalent of the acid content in 50 μ L of the choline-citronellic acid ionic liquid, the blood glucose levels only dropped to about 30% of starting levels, a level similar to that observed following a 10 μ L ionic liquid dose. Citronellic acid alone does not provide the same efficacy in lowering blood glucose as the choline-citronellic acid ionic liquid.

Example 8: Reduction in Blood Glucose Levels Following Oral and Subcutaneous Choline-Citronellic Acid Administration

[0128] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed with choline-citronellic acid via either subcutaneous injection or liquid gavage. A fasting control group was dosed orally with a saline solution. About 250 μ L of blood was collected at regular intervals in order to determine the blood glucose level. The obtained values, plotted as mean percent change $\pm$ standard error in blood glucose levels with respect to initial reading (n=4) versus time, are shown in **FIG. 10**.

[0129] Both oral and subcutaneous delivery of choline-citronellic acid resulted in reduced blood glucose levels in rats. In both cases, the glucose level dropped within the first two hours and increased over time.

Example 9: Reduction in Blood Glucose Levels Following Oral Choline-Citronellic Acid Administration in a Rat Model of Type 1 Diabetes

[0130] Adult Streptozocin-induced diabetic male Wistar rats and non-diabetic rats were fasted overnight but given free access to water and subsequently orally dosed with choline-citronellic acid via liquid gavage. About 250 μ L of blood was collected at regular intervals in order to determine the blood glucose level. The obtained values, plotted as mean percent change $\pm$ standard error in blood glucose levels with respect to initial reading (n=4) versus time, are shown in **FIG. 11**.

[0131] Oral delivery of choline-citronellic acid produced a substantial drop in blood glucose levels. In a healthy rat, the glucose level dropped and recovered slowly with time. In the Type 1 diabetes model, on the other hand, the glucose level continued to drop after oral administration of choline-citronellic acid, demonstrating the potential of choline-citronellic acid as a therapeutic for Type 1 diabetes.

Example 10: Induced Insulation Secretion Following Choline-Citronellic Acid Administration in a Rat Model of Type 1 Diabetes

[0132] Adult Streptozotocin-induced diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed with choline-citronellic acid via subcutaneous injection

(SC), oral liquid gavage, or jejunum placement via a catheter (Jejunum catheter). A fasting control group was not treated. About 250 μ L of blood was collected at regular intervals in order to determine the insulin serum concentration. The obtained values, plotted as mean plasma insulin concentration $\pm$ standard error (n=3) versus time, are shown in **FIG. 12**.

[0133] Stimulation of insulin secretion was observed for all administration methods of choline-citronellic acid, demonstrating the potential of choline-citronellic acid as a therapeutic for Type 1 diabetes patients who lack the natural ability to produce insulin in the pancreas. Choline-citronellic acid could be particularly useful for newly diagnosed Type 1 diabetes patients or prediabetic patients, for whom the pancreas still maintains some insulin-producing cells which can be stimulated by choline-citronellic acid administration to produce insulin.

Example 11: Increased Glucose Excretion in Urine Following Choline-Citronellic Acid Administration

[0134] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed via intrajejunal injection with choline-citronellic acid at varying doses and monitored for blood glucose. At 1.5 hours post-injection, urine was sampled from the bladder to determine glucose concentration. The obtained glucose concentration values, plotted as percent change in blood glucose levels with respect to initial reading versus dose level and change in glucose concentration in the urine collected from the bladder versus dose level (n=1), are shown in **FIG. 13**.

[0135] As evidenced by the declining blood glucose levels and increasing glucose concentration in the urine, choline-citronellic acid reduced the ability of the kidneys to reabsorb glucose, enhancing the amount of glucose removal from the body and helping to reduce blood glucose levels.

Example 12: Delivery of Liraglutide With Choline-Hydrocinnamic Acid

[0136] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed via intrajejunal injection with liraglutide and choline-hydrocinnamic acid. A fasting control group was dosed with a saline solution via intrajejunal injection. About 250 μ L of blood was collected at hourly intervals in order to determine the liraglutide serum concentration. The obtained values, plotted as mean liraglutide serum concentration $\pm$ standard error (n=3) in ng/mL versus time, are shown in **FIG. 14**.

[0137] When delivered as a choline-hydrocinnamic acid formulation, liraglutide was delivered into blood circulation with surprisingly high serum concentration. Oral delivery of liraglutide is known to be difficult. For example, Buckley and coworkers demonstrated only minute absorption of

liraglutide even in the presence of well-known permeation enhancers (*Sci. Transl. Med.* **2018**, *10*, eaar7047). The blood concentrations of liraglutide reported herein are approximately 4,400-fold greater than those reported in the literature. This unexpected level of liraglutide delivery demonstrates the promise of choline-hydrocinnamic acid as a diabetes therapeutic, especially for the treatment of Type 2 diabetes, for which the therapeutic benefits of liraglutide are well established. However, the fact that the current standard of care for liraglutide is daily injections poses a significant hurdle in compliance and patient acceptance. An oral pill that can deliver liraglutide would dramatically improve patient impact.

Example 13: Delivery of Liraglutide With Various Choline-Based Ionic Liquids

[0138] Adult non-diabetic male Wistar rats were fasted overnight but given free access to water and subsequently dosed via intrajejunal injection with liraglutide and one of various choline-based ionic liquids. The peak 5-hour liraglutide concentrations obtained for the various ionic liquids are summarized in **Table 2**:

Table 2: Peak 5-Hour Concentration of Liraglutide Delivered Via Intrajejunal Injection with Various Choline-Based Ionic Liquids

Ionic Liquid	Peak 5-Hour Liraglutide Concentration (ng/mL)
Choline-Hydrocinnamic acid	2086
Choline-Cinnamic acid	2057
Choline-Glutaric acid	365
Choline-Malonic acid	234
Choline-Octenoic acid	201
Choline-Linoleic acid	44
Choline-Citronellic acid	4.5

[0139] The amount of liraglutide delivered depended on the composition of the ionic liquid. Choline-citronellic acid yielded a modest but significant absorption to yield a peak concentration of 4.5 ng/mL. Choline-linoleic acid improved the concentration by about 10-fold to 44 ng/mL. Choline-malonic acid further improved the absorption to yield a blood liraglutide concentration of 365 ng/mL. Unexpectedly, choline-hydrocinnamic acid yielded a blood liraglutide concentration of greater than 2000 ng/mL, a 500-fold improvement over choline-citronellic acid.

Example 14: Lira-C-Cinnamic 1:1/dogs - delivery of Liraglutide With Choline-Cinnamic Acid 1:1 to the stomach or duodenum

[0140] Adult non-diabetic male Beagle dogs were fasted overnight but given free access to water and subsequently dosed under anesthesia via endoscopic placement to the stomach (as a liquid or

capsule) or duodenum (as a liquid) with 0.6 mg/kg Liraglutide with Choline-Cinnamic Acid 1:1. Dogs were recovered and plasma collected over a 12h period. Control groups included intravenous (IV, 0.03 mg/kg) and subcutaneous (SC, 0.06 mg/kg) dosing. 0.6 mg/kg unformulated Liraglutide was also administered to the stomach endoscopically (FIG. 15).

Example 15: Exenatide-C-Cinnamic 1:1/dogs - delivery of Exenatide With Choline-Cinnamic Acid 1:1 to the stomach

[0141] Adult non-diabetic male Beagle dogs were fasted overnight but given free access to water and subsequently dosed under anesthesia via endoscopic placement to the stomach with 0.6 mg/kg Exenatide with Choline-Cinnamic Acid 1:1 in liquid form (Exenatide-IL). Dogs were recovered and plasma collected over a 12h period. Control groups included intravenous (IV, 0.03 mg/kg) and subcutaneous (SC, 0.06 mg/kg) dosing. Unformulated exenatide was also administered as a liquid in buffer (saline, 0.6 mg/kg) to the stomach endoscopically (FIG. 16).

Example 16: Semaglutide-C-Cinnamic 1:1 capsules/dogs - delivery of Semaglutide With Choline-Cinnamic Acid 1:1 to the stomach in gelatin and HPMC capsules

[0142] Adult non-diabetic male Beagle dogs were fasted overnight but given free access to water and subsequently dosed via endoscopic placement under anesthesia to the stomach 0.6 mg/kg Semaglutide with Choline-Cinnamic Acid 1:1 contained in 0, 00 or 000 gelatin capsules coated with Evonik's EPO coating or 0 HPMC capsule. Dogs were recovered and plasma collected over a 12h period (FIG. 17).

Example 17: Semaglutide-C-Cinnamic 1:1/dogs - delivery of Semaglutide With Choline-Cinnamic Acid 1:1 to the stomach

[0143] Adult non-diabetic male Beagle dogs were fasted overnight but given free access to water and subsequently dosed under anesthesia via endoscopic placement to the stomach with 0.6 mg/kg Semaglutide with Choline-Cinnamic Acid 1:1, either in liquid form (Sema-IL), or in a gelatin capsule (00 Gel Capsule). Dogs were recovered and plasma collected over a 12h period. Control groups included intravenous (IV, 0.03 mg/kg) and subcutaneous (SC, 0.06 mg/kg) dosing. Unformulated semaglutide was also administered as a liquid in buffer (PPB, 0.6 mg/kg) to the stomach endoscopically. As a comparator, 0.6 mg/kg semaglutide was mixed with SNAC, a permeation enhancer and dosed via liquid to the stomach endoscopically (FIG. 18).

Example 18: mixture of Liraglutide/Exenatide - co-delivery of Liraglutide and Exenatide With Choline-Cinnamic Acid 1:1

[0144] Adult non-diabetic male Beagle dogs were fasted overnight but given free access to water and subsequently dosed under anesthesia via endoscopic placement to the stomach with a mixture of 0.3 mg/kg each Liraglutide and Exenatide with Choline-Cinnamic Acid 1:1 in liquid form. Dogs were recovered and plasma collected over a 12h period. (FIG. 19).

Example 19: safety/tox - safety profile of Choline-Cinnamic Acid 1:1.5

[0145] Adult non-diabetic male Wistar rats were dosed daily for 30 days via oral gavage with either 25 or 100 uL Choline-Cinnamic Acid 1:1.5. Two placebo groups received either 25 or 100 uL saline. Animals were observed daily for general health and weight recorded. On day 31, all animals were euthanized, and tissue, blood and plasma samples collected for analysis. Sections of organs (heart, lungs, liver, kidneys, and spleen) and all GI tract sections (stomach, duodenum, jejunum, ileum and colon) were stained with H&E and an expert pathologist read the results, concluding that there was no significant difference found in their histological examination of IL or saline (placebo) dosed animals' organs and GI tract tissues (FIG. 20). Blood cell counts were not statistically significantly different between the IL-dosed and placebo groups for either dose level, nor were plasma indicators of organ toxicity (FIG. 21). IHC staining of tight junction proteins Claudin-1 and Occludin of the GI tract tissues (duodenum, jejunum, ileum and colon) showed no difference between the IL-dosed and placebo groups for either dose level (FIG. 22). There was no difference in overall health or body weight gain between the IL-dosed and placebo groups (FIG. 23).

Example 20: Delivery of drugs formulated with Choline-Cinnamic Acid in various cation:anion ratios to the stomach or duodenum

[0146] Adult non-diabetic male Beagle dogs are fasted overnight but given free access to water and subsequently dosed under anesthesia via endoscopic placement to the stomach (as a liquid or capsule (e.g., gelatin capsule)) or duodenum (as a liquid) with a drug, for example, Liraglutide, Exenatide, or Semaglutide, formulated with Choline-Cinnamic Acid in a ratio of, for example, from about 4:1 to about 1:4. For instance, Choline-Cinnamic Acid is formulated in any one of the ratio as described in paragraphs [0085] or [0087]. Dogs are recovered and plasma is collected over a 12h period. Control groups include intravenous (IV) and subcutaneous (SC) dosing. The same dose of the unformulated drug, for example, Liraglutide, Exenatide, or Semaglutide, is also administered to the stomach endoscopically.

Example 21: Delivery of drugs formulated with various ionic liquids to the stomach or duodenum

[0147] Adult non-diabetic male Beagle dogs are fasted overnight but given free access to water and subsequently dosed under anesthesia via endoscopic placement to the stomach (as a liquid or capsule (e.g., gelatin capsule)) or duodenum (as a liquid) with a drug, for example, Liraglutide, Exenatide, or Semaglutide, formulated with Choline-Hydrocinnamic acid, Choline-Glutaric acid, Choline-Malonic acid, Choline-Octenoic acid, or Choline-Citronellic acid in a ratio of, for example, from about 4:1 to about 1:4. For instance, Choline-Hydrocinnamic acid, Choline-Glutaric acid, Choline-Malonic acid, Choline-Octenoic acid, or Choline-Citronellic acid is formulated at any of the ratio as described in paragraphs [0085] or [0087]. Dogs are recovered and plasma is collected over a 12h period. Control groups include intravenous (IV) and subcutaneous (SC) dosing. The same dose of the unformulated drug, for example, Liraglutide, Exenatide, or Semaglutide, is also administered to the stomach endoscopically.

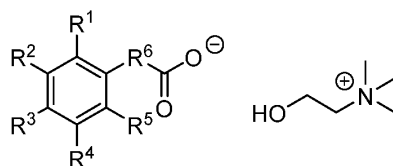
[0148] While preferred embodiments of the present invention have been shown and described herein, it will be obvious to those skilled in the art that such embodiments are provided by way of example only. Numerous variations, changes, and substitutions will now occur to those skilled in the art without departing from the invention. It should be understood that various alternatives to the embodiments of the invention described herein may be employed in practicing the invention. It is intended that the following claims define the scope of the invention and that methods and structures within the scope of these claims and their equivalents be covered thereby.

CLAIMS

WHAT IS CLAIMED IS:

1. A method of treating a disease or disorder in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of a composition comprising an ionic liquid.
2. The method of claim 1, wherein the disease or disorder is diabetes.
3. The method of claim 1 or claim 2, wherein the disease or disorder is Type 1 diabetes.
4. The method of claim 1 or claim 2, wherein the disease or disorder is Type 2 diabetes.
5. The method of claim 1, wherein the disease or disorder is non-alcoholic steatohepatitis.
6. A method of treating obesity, preventing weight gain, or reducing weight in a subject in need thereof, comprising administering to the subject a therapeutically effective amount of a composition comprising an ionic liquid.
7. The method of any one of claims 1 to 6, wherein the composition is administered via subcutaneous, intravenous, or oral administration.
8. The method of any one of claims 1 to 7, wherein the composition is administered via oral administration.
9. The method of any one of claims 1 to 8, wherein the composition is administered as a liquid-filled capsule.
10. The method of any one of claims 1 to 9, wherein the composition is administered in a single dose.
11. The method of any one of claims 1 to 9, wherein the composition is administered in multiple doses.
12. The method of any one of claims 1 to 11, wherein the composition is administered to a mucus membrane.
13. The method of any one of claims 1 to 12, wherein the composition comprises the ionic liquid at a concentration of at least 0.1% weight per volume.
14. The method of any one of claims 1 to 12, wherein the composition comprises the ionic liquid at a concentration of at least 0.05M.
15. The method of any one of claims 1 to 14, wherein the ionic liquid comprises a cation:anion ratio of from about 4:1 to about 1:4.

16. The method of any one of claims 1 to 15, wherein the ionic liquid is represented by Formula (I):



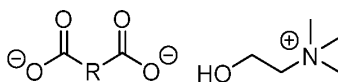
Formula (I),

wherein:

R¹, R², R³, R⁴, and R⁵ are independently selected from hydrogen, halo, cyano, nitro, amino, C₁₋₆alkoxy, C₁₋₆heteroalkyl, C₁₋₆haloalkyl, C₁₋₆alkyl, C₂₋₆alkenyl, and C₂₋₆alkynyl; and

R⁶ is selected from C₁₋₆alkyl, C₂₋₆alkenyl, and C₂₋₆alkynyl.

17. The method of claim 16, wherein at least two of R¹, R², R³, R⁴, and R⁵ are hydrogen.
18. The method of claim 17, wherein at least three of R¹, R², R³, R⁴, and R⁵ are hydrogen.
19. The method of claim 18, wherein R¹, R², R³, R⁴, and R⁵ are hydrogen.
20. The method of claim 16, wherein R⁶ is selected from C₁₋₆alkyl and C₂₋₆alkenyl.
21. The method of claim 20, wherein R⁶ is C₁₋₆alkyl.
22. The method of claim 21, wherein R⁶ is C₂alkyl.
23. The method of claim 20, wherein R⁶ is C₁₋₆alkenyl.
24. The method of claim 23, wherein R⁶ is C₂alkenyl.
25. The method of any one of claims 1 to 15, wherein the ionic liquid is represented by Formula (II):



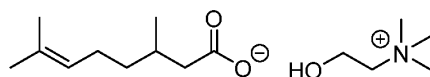
Formula (II),

wherein:

R is selected from C₁₋₆alkyl, C₂₋₆alkenyl, and C₂₋₆alkynyl.

26. The method of claim 25, wherein R is C₁₋₆alkyl.
27. The method of claim 26, wherein R is C₁alkyl.

28. The method of claim 26, wherein R is C₃alkyl.
29. The method of any one of claims 1 to 15, wherein the ionic liquid is represented by Formula (III):



Formula (III).

30. The method of any one of claims 1 to 15, wherein the ionic liquid comprises a cholinium cation and an anion selected from cinnamate, hydrocinnamate, malonate, citronellate, and glutarate.
31. The method of claim 30, wherein the anion is selected from cinnamate, hydrocinnamate, and citronellate.
32. The method of any one of claims 1 to 31, wherein the composition further comprises one or more additional agents.
33. The method of claim 32, wherein the one or more additional agents are selected from a nucleic acid, a small molecule, and a polypeptide.
34. The method of claim 33, wherein the one or more additional agents comprise a nucleic acid.
35. The method of claim 33, wherein the one or more additional agents comprise a small molecule.
36. The method of claim 33, wherein the one or more additional agents comprise a polypeptide.
37. The method of claim 32, wherein the one or more additional agents are selected from a glucagon-like peptide (GLP-1), a glucagon-like peptide derivative, and a glucagon-like peptide mimetic.
38. The method of claim 37, wherein the one or more additional agents are selected from liraglutide, exenatide, and semaglutide.
39. The method of claim 38, wherein the one or more additional agents comprise liraglutide.
40. The method of claim 32, wherein the one or more additional agents are selected from insulin and pramlintide.
41. The method of any one of claims 1 to 40, wherein the composition further comprises a pharmaceutically acceptable excipient.

42. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and hydrocinnamic acid in a 1:2 molar ratio.
43. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and cinnamic acid in a 1:1 molar ratio.
44. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and glutaric acid in a 1:1 molar ratio.
45. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and malonic acid in a 1:1 molar ratio.
46. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and octenoic acid in a 1:1 molar ratio.
47. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and octenoic acid in a 1:2 molar ratio.
48. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and citronellic acid in a 1:1 molar ratio.
49. A composition comprising an ionic liquid, wherein the ionic liquid comprises choline and citronellic acid in a 1:2 molar ratio.
50. The composition of any one of claims 42 to 49, further comprising one or more additional agents.
51. The composition of claim 50, wherein the one or more additional agents are selected from a nucleic acid, a small molecule, and a polypeptide
52. The composition of claim 51, wherein the one or more additional agents comprise a nucleic acid.
53. The composition of claim 51, wherein the one or more additional agents comprise a small molecule.
54. The composition of claim 51, wherein the one or more additional agents comprise a polypeptide.
55. The composition of claim 50, wherein the one or more additional agents are selected from a glucagon-like peptide (GLP-1), a glucagon-like peptide derivative, and a glucagon-like peptide mimetic.

56. The composition of claim 55, wherein the one or more additional agents are selected from liraglutide, exenatide, and semaglutide.
57. The composition of claim 56, wherein the one or more additional agents comprise liraglutide.
58. The composition of claim 50, wherein the one or more additional agents are selected from insulin and pramlintide.
59. A pharmaceutical composition comprising the composition of any one of claims 42 to 49 and a pharmaceutically acceptable excipient.