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(57) Abstract: Among other things, the present disclosure provides technologies including 1-66 formulations, compositions, methods, etc. for treating various cancers.



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STAPLED PEPTIDES AND METHODS THEREOF

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to United States Provisional Application Nos. 63/471,492, filed June 6, 2023, and 63/510,357, filed June 26, 2023, the entirety of each of which is incorporated herein by reference.

BACKGROUND

[0002] Stapled peptides are useful for various applications. For example, as biologically active agents, they can be utilized to modulate various biological functions.

SUMMARY

[0003] I-66 is a synthetic, helically constrained polypeptide. Various technologies have confirmed that it binds directly to an oncogenic protein, beta-catenin, and its binding to beta-catenin leads to blockade of beta-catenin:TCF (T-cell factor) complex formation, which has been confirmed in various in vitro and in vivo assays, including PDX mouse models, to inhibit downstream transcriptional program driven by the complex and dramatically inhibiting tumor growth.

[0004] Among other things, the present disclosure provides technologies, e.g., formulations, doses, dosage regimen, etc., for treating conditions, disorders or diseases particularly certain types of cancer. In some embodiments, the present disclosure provides high purity I-66 drug substance. In some embodiments, the present disclosure provides I-66 drug products. In some embodiments, the present disclosure provides I-66 formulations which are liquid compositions in which I-66 has sufficiently high solubility and stability for clinical applications.

[0005] The present disclosure also encompasses description of useful clinical development programs for I-66 to demonstrate and confirm that it can be utilized as described herein to treat conditions, disorders or diseases including cancer. In some embodiments, clinical programs include an open-label, in-human phase 1/2, multi-center study to evaluate and confirm the safety, pharmacokinetics (PK), pharmacodynamics (PD) and antitumor activity of I-66. Among other things, objectives of clinical studies include to demonstrate the safety and tolerability profile of DS-1 and to assess the antitumor activity of DS-1 in cancer patients.

[0006] In some embodiments, the present disclosure provides methods comprising administering or delivering to a subject an effective amount of I-66, wherein the subject is suffering from cancer. In some embodiments, I-66 is administered or delivered as a pharmaceutically acceptable salt. In some embodiments, I-66 is administered or delivered via a pharmaceutical composition described herein. In some embodiments, I-66 is administered or delivered via a formulation as described herein. In some embodiments, a subject is a patient with advanced or metastatic microsatellite stable (MSS) colorectal cancer (CRC). In some embodiments, a subject is a patient with a solid tumor with a WNT-pathway activating mutation (WPAM).

[0007] In some embodiments, the present disclosure provides pharmaceutical compositions comprising or delivering I-66 and a pharmaceutical acceptable carrier. In some embodiments, I-66 is in a pharmaceutically acceptable salt form. In some embodiments, a provided composition comprises a pharmaceutically acceptable salt form of I-66. In some embodiments, in various compositions and methods, I-66 is provided as one or more pharmaceutically acceptable salt forms.

[0008] In some embodiments, the present disclosure provides methods for modulating a property, activity and/or function of beta-catenin, comprising contacting beta-catenin with I-66. In some embodiments, the present disclosure provides methods for modulating a property, activity and/or function of beta-catenin in a system comprising beta-catenin, comprising administering to a system an effective amount of I-66. In some embodiments, the present disclosure provides methods for modulating a property, activity and/or function of beta-catenin in a system expressing beta-catenin, comprising administering or delivering to a system an effective amount of I-66. In some embodiments, an activity of beta-catenin is inhibited or reduced. In some embodiments, a function of beta-catenin is inhibited or reduced. In some embodiments, a property, activity and/or function is associated with beta-catenin/TCF interaction.

[0009] In some embodiments, the present disclosure provides methods for modulating beta-catenin/TCF interaction. In some embodiments, the present disclosure provides methods for modulating beta-catenin/TCF interaction, comprising contacting beta-catenin with I-66. In some embodiments, the present disclosure provides methods for modulating beta-catenin/TCF interaction in a system comprising beta-catenin and TCF, comprising administering or delivering to the system an effective amount of I-66. In some embodiments, the present disclosure provides methods for modulating beta-catenin/TCF interaction in a system expressing beta-catenin and TCF, comprising administering or delivering to the system an effective amount of I-66. In some embodiments, interactions between beta-catenin and TCF is reduced. In some embodiments, interactions between beta-catenin and TCF is inhibited.

[0010] In some embodiments, the present disclosure provides methods for inhibiting cell proliferation, comprising administering or delivering to a population of cells an effective amount of I-66. In some embodiments, the present disclosure provides methods for inhibiting cell proliferation in a system, comprising administering or delivering to the system an effective amount of I-66. In some embodiments, the present disclosure provides methods for inhibiting cell growth, comprising administering or delivering to a population of cells an effective amount of I-66. In some embodiments, the present disclosure provides methods for inhibiting cell growth in a system, comprising administering or delivering to the system an effective amount of I-66. In some embodiments, such cell proliferation is beta-catenin dependent. In some embodiments, such cell growth is beta-catenin dependent. In some embodiments, such proliferation or growth is dependent on beta-catenin interactions with TCF.

[0011] In some embodiments, the present disclosure provides methods for reducing or preventing activation of a WNT pathway. In some embodiments, the present disclosure provides methods for reducing or preventing activation of a WNT pathway in a system, comprising administering or delivering to the system

an effective amount of I-66.

[0012] In some embodiments, a system is *in vitro*. In some embodiments, a system is *ex vivo*. In some embodiments, a system is *in vivo*. In some embodiments, a system is or comprise a cell. In some embodiments, a system is or comprises a tissue. In some embodiments, a system is or comprises an organ. In some embodiments, a system is or comprises an organism. In some embodiments, a system is an animal. In some embodiments, a system is human. In some embodiments, a system is or comprises cells, tissues or organs associated with a condition, disorder or disease. In some embodiments, a system is or comprises cancer cells, e.g., colorectal cancer cells.

[0013] In some embodiments, I-66 is administered as pharmaceutically compositions that comprise or deliver I-66. In some embodiments, I-66 is provided and/or delivered in one or more pharmaceutically acceptable salt forms. In some embodiments, in a composition (e.g., a liquid composition of certain pH) I-66 may exist in various forms including various pharmaceutically acceptable salt forms.

[0014] Further description of certain embodiments of provided technologies is presented below.

BRIEF DESCRIPTION OF THE DRAWING

[0015] Figure 1. ¹H NMR of a preparation of I-66 (DMSO-d₆, 373K).

[0016] Figure 2. Integration of peaks in a ¹H NMR spectrum of a preparation of I-66 prepared as described in Example 9 (DMSO-d₆, 373K). Those skilled in the art appreciate that integration may be further adjusted and/or optimized.

[0017] Figure 3. ¹H NMR of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 8. Receiver gain: 30.1. Relaxation Delay: 2.5000. Acquisition Time: 1.5139. Spectrometer Frequency: 900.30 MHz. Spectra Width: 10822.5. Lowest Frequency: -927.2. Acquired Size: 16384. Spectral Size: 65536. Digital Resolution: 0.17.

[0018] Figure 4. A region of ¹H NMR of a preparation of I-66. Top: 900 MHz ¹H of a preparation of I-66 in methanol-d₄, 298 K, about 10 mg/750 uL. Bottom: 500 MHz ¹H of a preparation of I-66 in methanol-d₄, 298 K, about 10 mg/500 uL. As shown here, the top spectra can provide improved resolution for various peaks.

[0019] Figure 5. A region of ¹H NMR of a preparation of I-66. Top: 900 MHz ¹H of a preparation of I-66 in methanol-d₄, 298 K, about 10 mg/750 uL. Bottom: 500 MHz ¹H of a preparation of I-66 in methanol-d₄, 298 K, about 10 mg/500 uL. As shown here, the top spectra can provide improved resolution for various peaks; certain NH peaks at about 7.7ppm that were observed in the 500 MHz data were absent in the 900 MHz data. As those skilled in the art appreciate, NH peaks may change depending on sample environments.

[0020] Figure 6. ¹³C NMR of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 8108. Receiver gain: 177.2. Relaxation Delay: 1.5000. Acquisition Time: 0.3277. Spectrometer Frequency: 226.40 MHz. Spectra Width: 50000.0. Lowest Frequency: -911.8. Acquired Size: 16384. Spectral Size: 65536. Digital Resolution: 0.76.

[0021] Figure 7. COSY of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 4. Receiver gain: 30.1. Relaxation Delay: 2.0000. Acquisition Time: 0.2272. Spectrometer Frequency: (900.30, 900.30). Spectra Width: (9014.4, 9009.0). Lowest Frequency: (-22.7, -23.3). Nucleus: (¹H, ¹H). Acquired Size: (2048, 1024). Spectral Size: (2048, 1024). Digital Resolution: (4.40, 8.80).

[0022] Figure 8. ¹H-¹³C HSQC of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 8. Receiver gain: 177.2. Relaxation Delay: 1.5000. Acquisition Time: 0.1137. Spectrometer Frequency: (900.30, 226.40). Spectra Width: (9009.0, 34013.6). Lowest Frequency: (-22.0, 288.5). Nucleus: (¹H, ¹³C). Acquired Size: (1024, 512). Spectral Size: (1024, 1024). Digital Resolution: (8.80, 33.22).

[0023] Figure 9. ¹H-¹³C HMBC of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K.

[0024] Figure 10. ¹H-¹H TOCSY of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 8. Receiver gain: 57.7. Relaxation Delay: 2.0000. Acquisition Time: 0.2272. Spectrometer Frequency: (900.30, 900.30). Spectra Width: (9014.4, 9009.0). Lowest Frequency: (-21.8, -23.3). Acquired Size: (2048, 800). Spectral Size: (2048, 1024). Digital Resolution: (4.40, 8.80).

[0025] Figure 11. ¹H-¹H NOESY of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 8. Receiver gain: 57.7. Relaxation Delay: 2.0000. Acquisition Time: 0.2272. Spectrometer Frequency: (900.30, 900.30). Spectra Width: (9014.4, 9009.0). Lowest Frequency: (-5.7, -3.0). Acquired Size: (2048, 1024). Spectral Size: (2048, 1024). Digital Resolution: (4.40, 8.80).

[0026] Figure 12. Expansion of ¹H-¹H NOESY of a preparation of I-66. Solvent: methanol-d₄. About 10 mg/750 uL. Temperature: 298 K. Number of scans: 8. Receiver gain: 57.7. Relaxation Delay: 2.0000. Acquisition Time: 0.2272. Spectrometer Frequency: (900.30, 900.30). Spectra Width: (9014.4, 9009.0). Lowest Frequency: (-5.7, -3.0). Acquired Size: (2048, 1024). Spectral Size: (2048, 1024). Digital Resolution: (4.40, 8.80). As shown herein, NOE peak between the olefin protons of the PL3-B5 staple (111 and 112) was observed. In some embodiments, this peak was stronger than the NOE peak between neighboring aromatic protons of 3Thi (20 and 21), and/or of the *p* and *m* protons in the phenyl group of Phe (48 and 47/49).

[0027] Figure 13. A study schematic. APC=adenomatous polyposis coli; CRC=colorectal cancer; GEJ=gastroesophageal junction; MSS=microsatellite stable; MTD=maximum tolerated dose; NSCLC=non-small cell lung cancer; PD=pharmacodynamics; RP2D=recommended Phase 2 dose; WPAM=Wnt pathway activating mutation.

[0028] Figure 14. Microfluidic Modulation Spectroscopy (MMS) data of an I-66 preparation. (A) Absolute absorbance spectrum of I-66 at 20 mg/mL in the Amide I band (shown 1588-1712 cm⁻¹). This

spectrum was averaged from triplicate measurements. (B) Second derivative plot of the absolute absorbance spectrum. The four peaks highlighted represent the main secondary structural motifs in this peptide molecule. (C) Relative abundance of each secondary structural motif in I-66.

DETAILED DESCRIPTION OF CERTAIN EMBODIMENTS

Definitions

[0029] As used herein, the following definitions shall apply unless otherwise indicated. For purposes of this disclosure, the chemical elements are identified in accordance with the Periodic Table of the Elements, CAS version, Handbook of Chemistry and Physics, 75th Ed. Additionally, general principles of organic chemistry are described in “Organic Chemistry”, Thomas Sorrell, University Science Books, Sausalito: 1999, and “March’s Advanced Organic Chemistry”, 5th Ed., Ed.: Smith, M.B. and March, J., John Wiley & Sons, New York: 2001.

[0030] *Administration*: As used herein, the term “administration” typically refers to the administration of a composition to a subject or system. Those of ordinary skill in the art will be aware of a variety of routes that may, in appropriate circumstances, be utilized for administration to a subject, for example a human. For example, in some embodiments, administration may be ocular, oral, parenteral, topical, *etc.* In some particular embodiments, administration may be bronchial (*e.g.*, by bronchial instillation), buccal, dermal (which may be or comprise, for example, one or more of topical to the dermis, intradermal, interdermal, transdermal, *etc.*), enteral, intra-arterial, intradermal, intragastric, intramedullary, intramuscular, intranasal, intraperitoneal, intrathecal, intravenous, intraventricular, within a specific organ (*e.g.*, intrahepatic), mucosal, nasal, oral, rectal, subcutaneous, sublingual, topical, tracheal (*e.g.*, by intratracheal instillation), vaginal, vitreal, *etc.* In some embodiments, administration may involve dosing that is intermittent (*e.g.*, a plurality of doses separated in time) and/or periodic (*e.g.*, individual doses separated by a common period of time) dosing. In some embodiments, administration may involve continuous dosing (*e.g.*, perfusion) for at least a selected period of time.

[0031] *Affinity*: As is known in the art, “affinity” is a measure of the tightness with a particular ligand (*e.g.*, an agent) binds to its partner (*e.g.*, beta-catenin or a portion thereof). Affinities can be measured in different ways. In some embodiments, affinity is measured by a quantitative assay. In some such embodiments, binding partner concentration may be fixed to be in excess of ligand concentration so as to mimic physiological conditions. Alternatively or additionally, in some embodiments, binding partner concentration and/or ligand concentration may be varied. In some such embodiments, affinity may be compared to a reference under comparable conditions (*e.g.*, concentrations).

[0032] *Agent*: In general, the term “agent”, as used herein, may be used to refer to a compound or entity of any chemical class including, for example, a polypeptide, nucleic acid, saccharide, lipid, small molecule, metal, or combination or complex thereof. In appropriate circumstances, as will be clear from context to those skilled in the art, the term may be utilized to refer to an entity that is or comprises a cell or organism, or

a fraction, extract, or component thereof. Alternatively or additionally, as context will make clear, the term may be used to refer to a natural product in that it is found in and/or is obtained from nature. In some instances, again as will be clear from context, the term may be used to refer to one or more entities that is man-made in that it is designed, engineered, and/or produced through action of the hand of man and/or is not found in nature. In some embodiments, an agent may be utilized in isolated or pure form; in some embodiments, an agent may be utilized in crude form. In some embodiments, potential agents may be provided as collections or libraries, for example that may be screened to identify or characterize active agents within them. In some cases, the term “agent” may refer to a compound or entity that is or comprises a polymer; in some cases, the term may refer to a compound or entity that comprises one or more polymeric moieties. In some embodiments, the term “agent” may refer to a compound or entity that is not a polymer and/or is substantially free of any polymer and/or of one or more particular polymeric moieties. In some embodiments, the term may refer to a compound or entity that lacks or is substantially free of any polymeric moiety. In some embodiments, an agent is a compound. In some embodiments, an agent is a stapled peptide.

[0033] *Amino acid*: In its broadest sense, as used herein, refers to any compound and/or substance that can be incorporated into a polypeptide chain, *e.g.*, through formation of one or more peptide bonds. In some embodiments, an amino acid comprising an amino group and an a carboxylic acid group. In some embodiments, an amino acid has the structure of $\text{NH}(\text{R}^{\text{a1}})-\text{L}^{\text{a1}}-\text{C}(\text{R}^{\text{a2}})(\text{R}^{\text{a3}})-\text{L}^{\text{a2}}-\text{COOH}$, wherein each variable is independently as described in the present disclosure. In some embodiments, an amino acid has the general structure $\text{NH}(\text{R}')-\text{C}(\text{R}')_2-\text{COOH}$, wherein each R' is independently as described in the present disclosure. In some embodiments, an amino acid has the general structure $\text{H}_2\text{N}-\text{C}(\text{R}')_2-\text{COOH}$, wherein R' is as described in the present disclosure. In some embodiments, an amino acid has the general structure $\text{H}_2\text{N}-\text{C}(\text{H})(\text{R}')-\text{COOH}$, wherein R' is as described in the present disclosure. In some embodiments, an amino acid is a naturally-occurring amino acid. In some embodiments, an amino acid is a non-natural amino acid; in some embodiments, an amino acid is a D-amino acid; in some embodiments, an amino acid is an L-amino acid. “Standard amino acid” refers to any of the twenty standard L-amino acids commonly found in naturally occurring peptides. “Nonstandard amino acid” refers to any amino acid, other than the standard amino acids, regardless of whether it is prepared synthetically or obtained from a natural source. In some embodiments, an amino acid, including a carboxy- and/or amino-terminal amino acid in a polypeptide, can contain a structural modification as compared with the general structure above. For example, in some embodiments, an amino acid may be modified by methylation, amidation, acetylation, pegylation, glycosylation, phosphorylation, and/or substitution (*e.g.*, of the amino group, the carboxylic acid group, one or more protons, one or more hydrogens, and/or the hydroxyl group) as compared with the general structure. In some embodiments, such modification may, for example, alter the circulating half-life of a polypeptide containing the modified amino acid as compared with one containing an otherwise identical unmodified amino acid. In some embodiments, such modification does not significantly alter a relevant activity of a polypeptide containing the modified amino acid, as compared with one containing an otherwise identical unmodified amino acid. As will be clear

from context, in some embodiments, the term “amino acid” may be used to refer to a free amino acid; in some embodiments it may be used to refer to an amino acid residue of a polypeptide.

[0034] *Analog:* As used herein, the term “analog” refers to a substance that shares one or more particular structural features, elements, components, or moieties with a reference substance. Typically, an “analog” shows significant structural similarity with the reference substance, for example sharing a core or consensus structure, but also differs in certain discrete ways. In some embodiments, an analog is a substance that can be generated from the reference substance, *e.g.*, by chemical manipulation of the reference substance. In some embodiments, an analog is a substance that can be generated through performance of a synthetic process substantially similar to (*e.g.*, sharing a plurality of steps with) one that generates the reference substance. In some embodiments, an analog is or can be generated through performance of a synthetic process different from that used to generate the reference substance.

[0035] *Animal:* As used herein refers to any member of the animal kingdom. In some embodiments, “*animal*” refers to humans, of either sex and at any stage of development. In some embodiments, “*animal*” refers to non-human animals, at any stage of development. In certain embodiments, the non-human animal is a mammal (*e.g.*, a rodent, a mouse, a rat, a rabbit, a monkey, a dog, a cat, a sheep, cattle, a primate, and/or a pig). In some embodiments, animals include, but are not limited to, mammals, birds, reptiles, amphibians, fish, insects, and/or worms. In some embodiments, an animal may be a transgenic animal, genetically engineered animal, and/or a clone.

[0036] *Approximately:* As used herein, the term “approximately” or “about,” as applied to one or more values of interest, refers to a value that is similar to a stated reference value. In certain embodiments, the term “approximately” or “about” refers to a range of values that fall within 25%, 20%, 19%, 18%, 17%, 16%, 15%, 14%, 13%, 12%, 11%, 10%, 9%, 8%, 7%, 6%, 5%, 4%, 3%, 2%, 1%, or less in either direction (greater than or less than) of the stated reference value unless otherwise stated or otherwise evident from the context (except where such number would exceed 100% of a possible value).

[0037] *Associated with:* Two events or entities are “associated” with one another, as that term is used herein, if the presence, level and/or form of one is correlated with that of the other. For example, a particular entity (*e.g.*, nucleic acid (*e.g.*, genomic DNA, transcripts, mRNA, *etc.*), polypeptide, genetic signature, metabolite, microbe, *etc.*) is considered to be associated with a particular disease, disorder, or condition, if its presence, level and/or form correlates with incidence of and/or susceptibility to the disease, disorder, or condition (*e.g.*, across a relevant population).

[0038] *Binding:* It will be understood that the term “binding”, as used herein, typically refers to a non-covalent association between or among agents. In many embodiments herein, binding is addressed with respect to particular agents and beta-catenin. It will be appreciated by those of ordinary skill in the art that such binding may be assessed in any of a variety of contexts. In some embodiments, binding is assessed with respect to beta-catenin. In some embodiments, binding is assessed with respect to one or more amino acid residues of beta-catenin. In some embodiments, binding is assessed with respect to one or more amino acid

residues corresponding to (e.g., similarly positioned in three dimensional space and/or having certain similar properties and/or functions) those of beta-catenin.

[0039] *Binding site:* The term “binding site”, as used herein, refers to a region of a target polypeptide, formed in three-dimensional space, that includes one or more or all interaction residues of the target polypeptide. In some embodiments, “binding site” may refer to one or more amino acid residues which comprise or are one or more or all interaction amino acid residues of a target polypeptide. As will be understood by those of ordinary skill in the art, a binding site may include residues that are adjacent to one another on a linear chain, and/or that are distal to one another on a linear chain but near to one another in three-dimensional space when a target polypeptide is folded. A binding site may comprise amino acid residues and/or saccharide residues.

[0040] *Carrier:* as used herein, refers to a diluent, adjuvant, excipient, or vehicle with which a composition is administered. In some exemplary embodiments, carriers can include sterile liquids, such as, for example, water and oils, including oils of petroleum, animal, vegetable or synthetic origin, such as, for example, peanut oil, soybean oil, mineral oil, sesame oil and the like. In some embodiments, carriers are or include one or more solid components.

[0041] *Comparable:* As used herein, the term “comparable” refers to two or more agents, entities, situations, sets of conditions, *etc.*, that may not be identical to one another but that are sufficiently similar to permit comparison there between so that one skilled in the art will appreciate that conclusions may reasonably be drawn based on differences or similarities observed. In some embodiments, comparable sets of conditions, circumstances, individuals, or populations are characterized by a plurality of substantially identical features and one or a small number of varied features. Those of ordinary skill in the art will understand, in context, what degree of identity is required in any given circumstance for two or more such agents, entities, situations, sets of conditions, *etc.* to be considered comparable. For example, those of ordinary skill in the art will appreciate that sets of circumstances, individuals, or populations are comparable to one another when characterized by a sufficient number and type of substantially identical features to warrant a reasonable conclusion that differences in results obtained or phenomena observed under or with different sets of circumstances, individuals, or populations are caused by or indicative of the variation in those features that are varied.

[0042] *Derivative:* As used herein, the term “derivative” refers to a structural analogue of a reference substance. That is, a “derivative” is a substance that shows significant structural similarity with the reference substance, for example sharing a core or consensus structure, but also differs in certain discrete ways. In some embodiments, a derivative is a substance that can be generated from the reference substance by chemical manipulation. In some embodiments, a derivative is a substance that can be generated through performance of a synthetic process substantially similar to (e.g., sharing a plurality of steps with) one that generates the reference substance.

[0043] *Dosage form or unit dosage form:* Those skilled in the art will appreciate that the term “dosage

form” may be used to refer to a physically discrete unit of an active agent (*e.g.*, a therapeutic or diagnostic agent) for administration to a subject. Typically, each such unit contains a predetermined quantity of active agent. In some embodiments, such quantity is a unit dosage amount (or a whole fraction thereof) appropriate for administration in accordance with a dosing regimen that has been determined to correlate with a desired or beneficial outcome when administered to a relevant population (*i.e.*, with a therapeutic dosing regimen). Those of ordinary skill in the art appreciate that the total amount of a therapeutic composition or agent administered to a particular subject is determined by one or more attending physicians and may involve administration of multiple dosage forms.

[0044] *Dosing regimen*: Those skilled in the art will appreciate that the term “dosing regimen” may be used to refer to a set of unit doses (typically more than one) that are administered individually to a subject, typically separated by periods of time. In some embodiments, a given therapeutic agent has a recommended dosing regimen, which may involve one or more doses. In some embodiments, a dosing regimen comprises a plurality of doses each of which is separated in time from other doses. In some embodiments, individual doses are separated from one another by a time period of the same length; in some embodiments, a dosing regimen comprises a plurality of doses and at least two different time periods separating individual doses. In some embodiments, all doses within a dosing regimen are of the same unit dose amount. In some embodiments, different doses within a dosing regimen are of different amounts. In some embodiments, a dosing regimen comprises a first dose in a first dose amount, followed by one or more additional doses in a second dose amount different from the first dose amount. In some embodiments, a dosing regimen comprises a first dose in a first dose amount, followed by one or more additional doses in a second dose amount same as the first dose amount. In some embodiments, a dosing regimen is correlated with a desired or beneficial outcome when administered across a relevant population (*i.e.*, is a therapeutic dosing regimen).

[0045] *“Improved,” “increased” or “reduced”*: As used herein, these terms, or grammatically comparable comparative terms, indicate values that are relative to a comparable reference measurement. For example, in some embodiments, an assessed value achieved with an agent of interest may be “improved” relative to that obtained with a comparable reference agent. Alternatively or additionally, in some embodiments, an assessed value achieved in a subject or system of interest may be “improved” relative to that obtained in the same subject or system under different conditions (*e.g.*, prior to or after an event such as administration of an agent of interest), or in a different, comparable subject (*e.g.*, in a comparable subject or system that differs from the subject or system of interest in presence of one or more indicators of a particular disease, disorder or condition of interest, or in prior exposure to a condition or agent, *etc.*). In some embodiments, comparative terms refer to statistically relevant differences (*e.g.*, that are of a prevalence and/or magnitude sufficient to achieve statistical relevance). Those skilled in the art will be aware, or will readily be able to determine, in a given context, a degree and/or prevalence of difference that is required or sufficient to achieve such statistical significance.

[0046] *Peptide*: The term “peptide” as used herein refers to a polypeptide. In some embodiments, a

peptide is a polypeptide that is relatively short, for example having a length of less than about 100 amino acids, less than about 50 amino acids, less than about 40 amino acids less than about 30 amino acids, less than about 25 amino acids, less than about 20 amino acids, less than about 15 amino acids, or less than 10 amino acids. In some embodiments, a length is about 5-20, 5-19, 5-18, 5-17, 5-16, 5-15, 10-20, 10-19, 10-18, 10-17, 10-16, 10-15, 11-20, 11-19, 11-18, 11-17, 11-16, 11-15, 12-20, 12-19, 12-18, 12-17, 12-16, 12-15, 13-20, 13-19, 13-18, 13-17, 13-16, 13-15, 14-20, 14-19, 14-18, 14-17, 14-16, 14-15, or about 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, or 20 amino acids.

[0047] *Pharmaceutical composition:* As used herein, the term “pharmaceutical composition” refers to an active agent, formulated together with one or more pharmaceutically acceptable carriers. In some embodiments, active agent is present in unit dose amount appropriate for administration in a therapeutic regimen that shows a statistically significant probability of achieving a predetermined therapeutic effect when administered to a relevant population. In some embodiments, pharmaceutical compositions may be specially formulated for administration in solid or liquid form, including those adapted for the following: oral administration, for example, drenches (aqueous or non-aqueous solutions or suspensions), tablets, *e.g.*, those targeted for buccal, sublingual, and systemic absorption, boluses, powders, granules, pastes for application to the tongue; parenteral administration, for example, by subcutaneous, intramuscular, intravenous or epidural injection as, for example, a sterile solution or suspension, or sustained-release formulation; topical application, for example, as a cream, ointment, or a controlled-release patch or spray applied to the skin, lungs, or oral cavity; intravaginally or intrarectally, for example, as a pessary, cream, or foam; sublingually; ocularly; transdermally; or nasally, pulmonary, and to other mucosal surfaces.

[0048] *Pharmaceutically acceptable:* As used herein, the phrase “pharmaceutically acceptable” refers to those compounds, materials, compositions, 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.

[0049] *Pharmaceutically acceptable carrier:* As used herein, the term “pharmaceutically acceptable carrier” means a pharmaceutically-acceptable material, composition or vehicle, such as a liquid or solid filler, diluent, excipient, or solvent encapsulating material, involved in carrying or transporting the subject compound from one organ, or portion of the body, to another organ, or portion of the body. 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: sugars, such as lactose, glucose and sucrose; starches, such as corn starch and potato starch; cellulose, and its derivatives, such as sodium carboxymethyl cellulose, ethyl cellulose and cellulose acetate; powdered tragacanth; malt; gelatin; talc; excipients, such as cocoa butter and suppository waxes; oils, such as peanut oil, cottonseed oil, safflower oil, sesame oil, olive oil, corn oil and soybean oil; glycols, such as propylene glycol; polyols, such as glycerin, sorbitol, mannitol and polyethylene glycol; esters, such as ethyl

oleate and ethyl laurate; agar; buffering agents, such as magnesium hydroxide and aluminum hydroxide; alginic acid; pyrogen-free water; isotonic saline; Ringer's solution; ethyl alcohol; pH buffered solutions; polyesters, polycarbonates and/or polyanhydrides; and other non-toxic compatible substances employed in pharmaceutical formulations.

[0050] *Pharmaceutically acceptable salt*: The term "pharmaceutically acceptable salt", as used herein, refers to salts of such compounds that are appropriate for use in pharmaceutical contexts, i.e., salts which are, within the scope of sound medical judgment, suitable for use in contact with the tissues of humans and lower animals without undue toxicity, irritation, allergic response and the like, and are commensurate with a reasonable benefit/risk ratio. Pharmaceutically acceptable salts are well known. For example, S. M. Berge, et al. describes pharmaceutically acceptable salts in detail in J. Pharmaceutical Sciences, 66: 1-19 (1977). In some embodiments, pharmaceutically acceptable salts include, but are not limited to, nontoxic acid addition salts, which are salts of an amino group formed with inorganic acids such as hydrochloric acid, hydrobromic acid, phosphoric acid, sulfuric acid and perchloric acid or with organic acids such as acetic acid, maleic acid, tartaric acid, citric acid, succinic acid or malonic acid or by using other known methods such as ion exchange. In some embodiments, pharmaceutically acceptable salts include, but are not limited to, adipate, alginate, ascorbate, aspartate, benzenesulfonate, benzoate, bisulfate, borate, butyrate, camphorate, camphorsulfonate, citrate, cyclopentanepropionate, digluconate, dodecylsulfate, ethanesulfonate, formate, fumarate, glucoheptonate, glycerophosphate, gluconate, hemisulfate, heptanoate, hexanoate, hydroiodide, 2-hydroxyethanesulfonate, lactobionate, lactate, laurate, lauryl sulfate, malate, maleate, malonate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, nitrate, oleate, oxalate, palmitate, pamoate, pectinate, persulfate, 3-phenylpropionate, phosphate, picrate, pivalate, propionate, stearate, succinate, sulfate, tartrate, thiocyanate, *p*-toluenesulfonate, undecanoate, valerate salts, and the like. In some embodiments, pharmaceutically acceptable salts include, but are not limited to, nontoxic base addition salts, such as those formed by acidic groups of provided compounds with bases. Representative alkali or alkaline earth metal salts include salts of sodium, lithium, potassium, calcium, magnesium, and the like. In some embodiments, pharmaceutically acceptable salts are ammonium salts (e.g., $-N(R)_3^+$). In some embodiments, pharmaceutically acceptable salts are sodium salts. In some embodiments, pharmaceutically acceptable salts include, when appropriate, nontoxic ammonium, quaternary ammonium, and amine cations formed using counterions such as halide, hydroxide, carboxylate, sulfate, phosphate, nitrate, alkyl having from 1 to 6 carbon atoms, sulfonate and aryl sulfonate.

[0051] *Polypeptide*: As used herein refers to any polymeric chain of amino acids. In some embodiments, a polypeptide has an amino acid sequence that occurs in nature. In some embodiments, a polypeptide has an amino acid sequence that does not occur in nature. In some embodiments, a polypeptide has an amino acid sequence that is engineered in that it is designed and/or produced through action of the hand of man. In some embodiments, a polypeptide may comprise or consist of natural amino acids, non-natural amino acids, or both. In some embodiments, a polypeptide may comprise or consist of only natural

amino acids or only non-natural amino acids. In some embodiments, a polypeptide may comprise D-amino acids, L-amino acids, or both. In some embodiments, a polypeptide may comprise only D-amino acids. In some embodiments, a polypeptide may comprise only L-amino acids. In some embodiments, a polypeptide may include one or more pendant groups or other modifications, *e.g.*, modifying or attached to one or more amino acid side chains, at the polypeptide's N-terminus, at the polypeptide's C-terminus, or any combination thereof. In some embodiments, such pendant groups or modifications may be selected from the group consisting of acetylation, amidation, lipidation, methylation, pegylation, *etc.*, including combinations thereof. In some embodiments, a polypeptide may be cyclic, and/or may comprise a cyclic portion. In some embodiments, a polypeptide is not cyclic and/or does not comprise any cyclic portion. In some embodiments, a polypeptide is linear. In some embodiments, a polypeptide may be or comprise a stapled polypeptide. In some embodiments, the term "polypeptide" may be appended to a name of a reference polypeptide, activity, or structure; in such instances it is used herein to refer to polypeptides that share the relevant activity or structure and thus can be considered to be members of the same class or family of polypeptides. For each such class, the present specification provides and/or those skilled in the art will be aware of exemplary polypeptides within the class whose amino acid sequences and/or functions are known; in some embodiments, such exemplary polypeptides are reference polypeptides for the polypeptide class or family. In some embodiments, a member of a polypeptide class or family shows significant sequence homology or identity with, shares a common sequence motif (*e.g.*, a characteristic sequence element) with, and/or shares a common activity (in some embodiments at a comparable level or within a designated range) with a reference polypeptide of the class; in some embodiments with all polypeptides within the class). For example, in some embodiments, a member polypeptide shows an overall degree of sequence homology or identity with a reference polypeptide that is at least about 30-40%, and is often greater than about 50%, 60%, 70%, 80%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99% or more and/or includes at least one region (*e.g.*, a conserved region that may in some embodiments be or comprise a characteristic sequence element) that shows very high sequence identity, often greater than 90% or even 95%, 96%, 97%, 98%, or 99%. Such a conserved region usually encompasses at least 3-4 and often up to 20 or more amino acids; in some embodiments, a conserved region encompasses at least one stretch of at least 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15 or more contiguous amino acids. In some embodiments, a relevant polypeptide may comprise or consist of a fragment of a parent polypeptide. In some embodiments, a useful polypeptide as may comprise or consist of a plurality of fragments, each of which is found in the same parent polypeptide in a different spatial arrangement relative to one another than is found in the polypeptide of interest (*e.g.*, fragments that are directly linked in the parent may be spatially separated in the polypeptide of interest or vice versa, and/or fragments may be present in a different order in the polypeptide of interest than in the parent), so that the polypeptide of interest is a derivative of its parent polypeptide.

[0052] *Prevent or prevention:* as used herein when used in connection with the occurrence of a disease, disorder, and/or condition, refers to reducing the risk of developing the disease, disorder and/or condition

and/or to delaying onset of one or more characteristics or symptoms of the disease, disorder or condition. Prevention may be considered complete when onset of a disease, disorder or condition has been delayed for a predefined period of time.

[0053] *Protecting group*: The term “protecting group,” as used herein, is well known in the art and includes those described in detail in *Protecting Groups in Organic Synthesis*, T. W. Greene and P. G. M. Wuts, 3rd edition, John Wiley & Sons, 1999, the entirety of which is incorporated herein by reference. Also included are those protecting groups specially adapted for nucleoside and nucleotide chemistry described in *Current Protocols in Nucleic Acid Chemistry*, edited by Serge L. Beaucage et al. 06/2012, the entirety of Chapter 2 is incorporated herein by reference. Suitable amino-protecting groups include methyl carbamate, ethyl carbamate, 9-fluorenylmethyl carbamate (Fmoc), 9-(2-sulfo)fluorenylmethyl carbamate, 9-(2,7-dibromo)fluorenylmethyl carbamate, 2,7-di-*t*-butyl-[9-(10,10-dioxo-10,10,10,10-tetrahydrothioxanthyl)]methyl carbamate (DBD-Tmoc), 4-methoxyphenacyl carbamate (Phenoc), 2,2,2-trichloroethyl carbamate (Troc), 2-trimethylsilylethyl carbamate (Teoc), 2-phenylethyl carbamate (hZ), 1-(1-adamantyl)-1-methylethyl carbamate (Adpoc), 1,1-dimethyl-2-haloethyl carbamate, 1,1-dimethyl-2,2-dibromoethyl carbamate (DB-*t*-BOC), 1,1-dimethyl-2,2,2-trichloroethyl carbamate (TCBOC), 1-methyl-1-(4-biphenyl)ethyl carbamate (Bpoc), 1-(3,5-di-*t*-butylphenyl)-1-methylethyl carbamate (*t*-Bumeoc), 2-(2'- and 4'-pyridyl)ethyl carbamate (Pyoc), 2-(*N,N*-dicyclohexylcarboxamido)ethyl carbamate, *t*-butyl carbamate (BOC), 1-adamantyl carbamate (Adoc), vinyl carbamate (Voc), allyl carbamate (Alloc), 1-isopropylallyl carbamate (Ipaoc), cinnamyl carbamate (Coc), 4-nitrocinnamyl carbamate (Noc), 8-quinolyl carbamate, *N*-hydroxypiperidinyl carbamate, alkylthio carbamate, benzyl carbamate (Cbz), *p*-methoxybenzyl carbamate (Moz), *p*-nitrobenzyl carbamate, *p*-bromobenzyl carbamate, *p*-chlorobenzyl carbamate, 2,4-dichlorobenzyl carbamate, 4-methylsulfinylbenzyl carbamate (MsZ), 9-anthrylmethyl carbamate, diphenylmethyl carbamate, 2-methylthioethyl carbamate, 2-methylsulfonylethyl carbamate, 2-(*p*-toluenesulfonyl)ethyl carbamate, [2-(1,3-dithianyl)]methyl carbamate (Dmoc), 4-methylthiophenyl carbamate (Mtpc), 2,4-dimethylthiophenyl carbamate (Bmpc), 2-phosphonioethyl carbamate (Peoc), 2-triphenylphosphonioisopropyl carbamate (Ppoc), 1,1-dimethyl-2-cyanoethyl carbamate, *m*-chloro-*p*-acyloxybenzyl carbamate, *p*-(dihydroxyboryl)benzyl carbamate, 5-benzisoxazolylmethyl carbamate, 2-(trifluoromethyl)-6-chromonylmethyl carbamate (Troc), *m*-nitrophenyl carbamate, 3,5-dimethoxybenzyl carbamate, *o*-nitrobenzyl carbamate, 3,4-dimethoxy-6-nitrobenzyl carbamate, phenyl(*o*-nitrophenyl)methyl carbamate, phenothiazinyl-(10)-carbonyl derivative, *N'*-*p*-toluenesulfonylaminocarbonyl derivative, *N'*-phenylaminothiocarbonyl derivative, *t*-amyl carbamate, *S*-benzyl thiocarbamate, *p*-cyanobenzyl carbamate, cyclobutyl carbamate, cyclohexyl carbamate, cyclopentyl carbamate, cyclopropylmethyl carbamate, *p*-decyloxybenzyl carbamate, 2,2-dimethoxycarbonylvinyl carbamate, *o*-(*N,N*-dimethylcarboxamido)benzyl carbamate, 1,1-dimethyl-3-(*N,N*-dimethylcarboxamido)propyl carbamate, 1,1-dimethylpropynyl carbamate, di(2-pyridyl)methyl carbamate, 2-furanylmethyl carbamate, 2-iodoethyl carbamate, isoborynl carbamate, isobutyl carbamate, isonicotinyl carbamate, *p*-(*p'*-methoxyphenylazo)benzyl carbamate, 1-

methylcyclobutyl carbamate, 1-methylcyclohexyl carbamate, 1-methyl-1-cyclopropylmethyl carbamate, 1-methyl-1-(3,5-dimethoxyphenyl)ethyl carbamate, 1-methyl-1-(p-phenylazophenyl)ethyl carbamate, 1-methyl-1-phenylethyl carbamate, 1-methyl-1-(4-pyridyl)ethyl carbamate, phenyl carbamate, p-(phenylazo)benzyl carbamate, 2,4,6-tri-t-butylphenyl carbamate, 4-(trimethylammonium)benzyl carbamate, 2,4,6-trimethylbenzyl carbamate, formamide, acetamide, chloroacetamide, trichloroacetamide, trifluoroacetamide, phenylacetamide, 3-phenylpropanamide, picolinamide, 3-pyridylcarboxamide, N-benzoylphenylalanyl derivative, benzamide, p-phenylbenzamide, o-nitrophenylacetamide, o-nitrophenoxycetamide, acetoacetamide, (N'-dithiobenzoyloxycarbonylamino)acetamide, 3-(p-hydroxyphenyl)propanamide, 3-(o-nitrophenyl)propanamide, 2-methyl-2-(o-nitrophenoxycetamide), 2-methyl-2-(o-phenylazophenoxy)propanamide, 4-chlorobutanamide, 3-methyl-3-nitrobutanamide, o-nitrocinnamide, N-acetylmethionine derivative, o-nitrobenzamide, o-(benzoyloxymethyl)benzamide, 4,5-diphenyl-3-oxazolin-2-one, N-phthalimide, N-dithiasuccinimide (Dts), N-2,3-diphenylmaleimide, N-2,5-dimethylpyrrole, N-1,1,4,4-tetramethyldisilylazacyclopentane adduct (STABASE), 5-substituted 1,3-dimethyl-1,3,5-triazacyclohexan-2-one, 5-substituted 1,3-dibenzyl-1,3,5-triazacyclohexan-2-one, 1-substituted 3,5-dinitro-4-pyridone, N-methylamine, N-allylamine, N-[2-(trimethylsilyl)ethoxy]methylamine (SEM), N-3-acetoxypyrrolamine, N-(1-isopropyl-4-nitro-2-oxo-3-pyrroline-3-yl)amine, quaternary ammonium salts, N-benzylamine, N-di(4-methoxyphenyl)methylamine, N-5-dibenzosuberylamine, N-triphenylmethylamine (Tr), N-[(4-methoxyphenyl)diphenylmethyl]amine (MMTr), N-9-phenylfluorenylamine (PhF), N-2,7-dichloro-9-fluorenylmethylethylamine, N-ferrocenylmethylamino (Fcm), N-2-picolyllamino N'-oxide, N-1,1-dimethylthiomethylethylamine, N-benzylideneamine, N-p-methoxybenzylideneamine, N-diphenylmethylethylamine, N-[(2-pyridyl)mesityl]methylethylamine, N-(N', N'-dimethylaminomethylethyl)amine, N,N'-isopropylidenediamine, N-p-nitrobenzylideneamine, N-salicylideneamine, N-5-chlorosalicylideneamine, N-(5-chloro-2-hydroxyphenyl)phenylmethylethylamine, N-cyclohexylideneamine, N-(5,5-dimethyl-3-oxo-1-cyclohexenyl)amine, N-borane derivative, N-diphenylborinic acid derivative, N-[phenyl(pentacarbonylchromium- or tungsten)carbonyl]amine, N-copper chelate, N-zinc chelate, N-nitroamine, N-nitrosoamine, amine N-oxide, diphenylphosphinamide (Dpp), dimethylthiophosphinamide (Mpt), diphenylthiophosphinamide (Ppt), dialkyl phosphoramidates, dibenzyl phosphoramidate, diphenyl phosphoramidate, benzenesulfenamide, o-nitrobenzenesulfenamide (Nps), 2,4-dinitrobenzenesulfenamide, pentachlorobenzenesulfenamide, 2-nitro-4-methoxybenzenesulfenamide, triphenylmethylsulfenamide, 3-nitropyridinesulfenamide (Npys), p-toluenesulfonamide (Ts), benzenesulfonamide, 2,3,6-trimethyl-4-methoxybenzenesulfonamide (Mtr), 2,4,6-trimethoxybenzenesulfonamide (Mtb), 2,6-dimethyl-4-methoxybenzenesulfonamide (Pme), 2,3,5,6-tetramethyl-4-methoxybenzenesulfonamide (Mte), 4-methoxybenzenesulfonamide (Mbs), 2,4,6-trimethylbenzenesulfonamide (Mts), 2,6-dimethoxy-4-methylbenzenesulfonamide (iMds), 2,2,5,7,8-pentamethylchroman-6-sulfonamide (Pmc), methanesulfonamide (Ms), β -trimethylsilyl ethanesulfonamide (SES), 9-anthracenesulfonamide, 4-(4',8'-

dimethoxynaphthylmethyl)benzenesulfonamide (DNMBS), benzylsulfonamide, trifluoromethylsulfonamide, and phenacysulfonamide.

[0054] In some embodiments, suitable mono-protected amines include, but are not limited to, aralkylamines, carbamates, allyl amines, amides, and the like. Examples of suitable mono-protected amino moieties include t-butyloxycarbonylamino (–NHBOC), ethyloxycarbonylamino, methyloxycarbonylamino, trichloroethyloxycarbonylamino, allyloxycarbonylamino (–NHAlloc), benzyloxycarbonylamino (–NHCBZ), allylamino, benzylamino (–NHBn), fluorenylmethylcarbonyl (–NHFmoc), formamido, acetamido, chloroacetamido, dichloroacetamido, trichloroacetamido, phenylacetamido, trifluoroacetamido, benzamido, t-butylidiphenylsilyl, and the like. In some embodiments, suitable di-protected amines include amines that are substituted with two substituents independently selected from those described above as mono-protected amines, and further include cyclic imides, such as phthalimide, maleimide, succinimide, and the like. In some embodiments, suitable di-protected amines include pyrroles and the like, 2,2,5,5-tetramethyl-1,2,5]azadisilolidine and the like, and azide.

[0055] Suitably protected carboxylic acids further include, but are not limited to, silyl–, alkyl–, alkenyl–, aryl–, and arylalkyl–protected carboxylic acids. Examples of suitable silyl groups include trimethylsilyl, triethylsilyl, t-butyltrimethylsilyl, t-butylidiphenylsilyl, triisopropylsilyl, and the like. Examples of suitable alkyl groups include methyl, benzyl, p-methoxybenzyl, 3,4-dimethoxybenzyl, trityl, t-butyl, tetrahydropyran–2–yl. Examples of suitable alkenyl groups include allyl. Examples of suitable aryl groups include optionally substituted phenyl, biphenyl, or naphthyl. Examples of suitable arylalkyl groups include optionally substituted benzyl (e.g., p-methoxybenzyl (MPM), 3,4-dimethoxybenzyl, O-nitrobenzyl, p-nitrobenzyl, p-halobenzyl, 2,6-dichlorobenzyl, p-cyanobenzyl), and 2- and 4-picolyl. In some embodiments, suitable protected carboxylic acids include, but are not limited to, optionally substituted C₁₋₆ aliphatic esters, optionally substituted aryl esters, silyl esters, activated esters, amides, hydrazides, and the like. Examples of such ester groups include methyl, ethyl, propyl, isopropyl, butyl, isobutyl, benzyl, and phenyl ester, wherein each group is optionally substituted. Additional suitable protected carboxylic acids include oxazolines and ortho esters.

[0056] Suitable hydroxyl protecting groups include methyl, methoxymethyl (MOM), methylthiomethyl (MTM), t-butylthiomethyl, (phenyldimethylsilyl)methoxymethyl (SMOM), benzyloxymethyl (BOM), p-methoxybenzyloxymethyl (PMBM), (4-methoxyphenoxy)methyl (p-AOM), guaiacolmethyl (GUM), t-butoxymethyl, 4-pentenylloxymethyl (POM), siloxymethyl, 2-methoxyethoxymethyl (MEM), 2,2,2-trichloroethoxymethyl, bis(2-chloroethoxy)methyl, 2-(trimethylsilyl)ethoxymethyl (SEMOR), tetrahydropyranyl (THP), 3-bromotetrahydropyranyl, tetrahydrothiopyranyl, 1-methoxycyclohexyl, 4-methoxytetrahydropyranyl (MTHP), 4-methoxytetrahydrothiopyranyl, 4-methoxytetrahydrothiopyranyl S,S-dioxide, 1-[(2-chloro-4-methyl)phenyl]-4-methoxypiperidin-4-yl (CTMP), 1,4-dioxan-2-yl, tetrahydrofuran-2-yl, tetrahydrothiofuran-2-yl, 2,3,3a,4,5,6,7,7a-octahydro-7,8,8-trimethyl-4,7-methanobenzofuran-2-yl, 1-ethoxyethyl, 1-(2-chloroethoxy)ethyl, 1-methyl-1-methoxyethyl, 1-methyl-1-

benzyloxyethyl, 1-methyl-1-benzyloxy-2-fluoroethyl, 2,2,2-trichloroethyl, 2-trimethylsilylethyl, 2-(phenylselenyl)ethyl, t-butyl, allyl, p-chlorophenyl, p-methoxyphenyl, 2,4-dinitrophenyl, benzyl, p-methoxybenzyl, 3,4-dimethoxybenzyl, o-nitrobenzyl, p-nitrobenzyl, p-halobenzyl, 2,6-dichlorobenzyl, p-cyanobenzyl, p-phenylbenzyl, 2-picolyl, 4-picolyl, 3-methyl-2-picolyl N-oxido, diphenylmethyl, p,p'-dinitrobenzhydryl, 5-dibenzosuberyl, triphenylmethyl, α -naphthylidiphenylmethyl, p-methoxyphenyldiphenylmethyl, di(p-methoxyphenyl)phenylmethyl, tri(p-methoxyphenyl)methyl, 4-(4'-bromophenacyloxyphenyl)diphenylmethyl, 4,4',4''-tris(4,5-dichlorophthalimidophenyl)methyl, 4,4',4''-tris(levulinoyloxyphenyl)methyl, 4,4',4''-tris(benzoyloxyphenyl)methyl, 3-(imidazol-1-yl)bis(4',4''-dimethoxyphenyl)methyl, 1,1-bis(4-methoxyphenyl)-1'-pyrenylmethyl, 9-anthryl, 9-(9-phenyl)xanthenyl, 9-(9-phenyl-10-oxo)anthryl, 1,3-benzodithiolan-2-yl, benzisothiazolyl S,S-dioxido, trimethylsilyl (TMS), triethylsilyl (TES), triisopropylsilyl (TIPS), dimethylisopropylsilyl (IPDMS), diethylisopropylsilyl (DEIPS), dimethylthexylsilyl, t-butyl dimethylsilyl (TBDMS), t-butyl diphenylsilyl (TBDPS), tribenzylsilyl, tri-p-xylylsilyl, triphenylsilyl, diphenylmethylsilyl (DPMS), t-butyl methoxyphenylsilyl (TBMPS), formate, benzoylformate, acetate, chloroacetate, dichloroacetate, trichloroacetate, trifluoroacetate, methoxyacetate, triphenylmethoxyacetate, phenoxyacetate, p-chlorophenoxyacetate, 3-phenylpropionate, 4-oxopentanoate (levulinate), 4,4-(ethylenedithio)pentanoate (levulinoyldithioacetal), pivaloate, adamantate, crotonate, 4-methoxycrotonate, benzoate, p-phenylbenzoate, 2,4,6-trimethylbenzoate (mesitoate), alkyl methyl carbonate, 9-fluorenylmethyl carbonate (Fmoc), alkyl ethyl carbonate, alkyl 2,2,2-trichloroethyl carbonate (Troc), 2-(trimethylsilyl)ethyl carbonate (TMSEC), 2-(phenylsulfonyl) ethyl carbonate (Psec), 2-(triphenylphosphonio) ethyl carbonate (Peoc), alkyl isobutyl carbonate, alkyl vinyl carbonate, alkyl allyl carbonate, alkyl p-nitrophenyl carbonate, alkyl benzyl carbonate, alkyl p-methoxybenzyl carbonate, alkyl 3,4-dimethoxybenzyl carbonate, alkyl o-nitrobenzyl carbonate, alkyl p-nitrobenzyl carbonate, alkyl S-benzyl thiocarbonate, 4-ethoxy-1-naphthyl carbonate, methyl dithiocarbonate, 2-iodobenzoate, 4-azidobutyrate, 4-nitro-4-methylpentanoate, o-(dibromomethyl)benzoate, 2-formylbenzenesulfonate, 2-(methylthiomethoxy)ethyl, 4-(methylthiomethoxy)butyrate, 2-(methylthiomethoxymethyl)benzoate, 2,6-dichloro-4-methylphenoxyacetate, 2,6-dichloro-4-(1,1,3,3-tetramethylbutyl)phenoxyacetate, 2,4-bis(1,1-dimethylpropyl)phenoxyacetate, chlorodiphenylacetate, isobutyrate, monosuccinoate, (E)-2-methyl-2-butenate, o-(methoxycarbonyl)benzoate, α -naphthoate, nitrate, alkyl N,N,N',N'-tetramethylphosphorodiamidate, alkyl N-phenylcarbamate, borate, dimethylphosphinothioyl, alkyl 2,4-dinitrophenylsulfenate, sulfate, methanesulfonate (mesylate), benzylsulfonate, and tosylate (Ts). For protecting 1,2- or 1,3-diols, the protecting groups include methylene acetal, ethylidene acetal, 1-t-butylethylidene ketal, 1-phenylethylidene ketal, (4-methoxyphenyl)ethylidene acetal, 2,2,2-trichloroethylidene acetal, acetamide, cyclopentylidene ketal, cyclohexylidene ketal, cycloheptylidene ketal, benzylidene acetal, p-methoxybenzylidene acetal, 2,4-dimethoxybenzylidene ketal, 3,4-dimethoxybenzylidene acetal, 2-nitrobenzylidene acetal, methoxymethylene acetal, ethoxymethylene acetal, dimethoxymethylene ortho ester, 1-methoxyethylidene ortho ester, 1-ethoxyethylidene ortho ester, 1,2-

dimethoxyethylidene ortho ester, α -methoxybenzylidene ortho ester, 1-(N,N-dimethylamino)ethylidene derivative, α -(N,N'-dimethylamino)benzylidene derivative, 2-oxacyclopentylidene ortho ester, di-*t*-butylsilylene group (DTBS), 1,3-(1,1,3,3-tetraisopropylidisiloxanylidene) derivative (TIPDS), tetra-*t*-butoxydisiloxane-1,3-diylidene derivative (TBDS), cyclic carbonates, cyclic boronates, ethyl boronate, and phenyl boronate.

[0057] In some embodiments, a hydroxyl protecting group is acetyl, *t*-butyl, *t*butoxymethyl, methoxymethyl, tetrahydropyranyl, 1-ethoxyethyl, 1-(2-chloroethoxy)ethyl, 2-trimethylsilylethyl, *p*-chlorophenyl, 2,4-dinitrophenyl, benzyl, benzoyl, *p*-phenylbenzoyl, 2,6-dichlorobenzyl, diphenylmethyl, *p*-nitrobenzyl, triphenylmethyl (trityl), 4,4'-dimethoxytrityl, trimethylsilyl, triethylsilyl, *t*-butyldimethylsilyl, *t*-butyldiphenylsilyl, triphenylsilyl, triisopropylsilyl, benzoylformate, chloroacetyl, trichloroacetyl, trifluoroacetyl, pivaloyl, 9-fluorenylmethyl carbonate, mesylate, tosylate, triflate, trityl, monomethoxytrityl (MMTr), 4,4'-dimethoxytrityl (DMTr) and 4,4',4''-trimethoxytrityl (TMTr), 2-cyanoethyl (CE or Cne), 2-(trimethylsilyl)ethyl (TSE), 2-(2-nitrophenyl)ethyl, 2-(4-cyanophenyl)ethyl 2-(4-nitrophenyl)ethyl (NPE), 2-(4-nitrophenylsulfonyl)ethyl, 3,5-dichlorophenyl, 2,4-dimethylphenyl, 2-nitrophenyl, 4-nitrophenyl, 2,4,6-trimethylphenyl, 2-(2-nitrophenyl)ethyl, butylthiocarbonyl, 4,4',4''-tris(benzoyloxy)trityl, diphenylcarbamoyl, levulinyl, 2-(dibromomethyl)benzoyl (Dbmb), 2-(isopropylthiomethoxymethyl)benzoyl (Ptmt), 9-phenylxanthene-9-yl (pixyl) or 9-(*p*-methoxyphenyl)xanthine-9-yl (MOX). In some embodiments, each of the hydroxyl protecting groups is, independently selected from acetyl, benzyl, *t*-butyldimethylsilyl, *t*-butyldiphenylsilyl and 4,4'-dimethoxytrityl. In some embodiments, the hydroxyl protecting group is selected from the group consisting of trityl, monomethoxytrityl and 4,4'-dimethoxytrityl group. In some embodiments, a phosphorous linkage protecting group is a group attached to the phosphorous linkage (e.g., an internucleotidic linkage) throughout oligonucleotide synthesis. In some embodiments, a protecting group is attached to a sulfur atom of an phosphorothioate group. In some embodiments, a protecting group is attached to an oxygen atom of an internucleotide phosphorothioate linkage. In some embodiments, a protecting group is attached to an oxygen atom of the internucleotide phosphate linkage. In some embodiments a protecting group is 2-cyanoethyl (CE or Cne), 2-trimethylsilylethyl, 2-nitroethyl, 2-sulfonylethyl, methyl, benzyl, *o*-nitrobenzyl, 2-(*p*-nitrophenyl)ethyl (NPE or Npe), 2-phenylethyl, 3-(*N*-tert-butylcarboxamido)-1-propyl, 4-oxopentyl, 4-methylthio-1-butyl, 2-cyano-1,1-dimethylethyl, 4-*N*-methylaminobutyl, 3-(2-pyridyl)-1-propyl, 2-[*N*-methyl-*N*-(2-pyridyl)]aminoethyl, 2-(*N*-formyl,*N*-methyl)aminoethyl, or 4-[*N*-methyl-*N*-(2,2,2-trifluoroacetyl)amino]butyl.

[0058] Protected thiols are well known in the art and include those described in detail in Greene (1999). Suitable protected thiols further include, but are not limited to, disulfides, thioethers, silyl thioethers, thioesters, thiocarbonates, and thiocarbamates, and the like. Examples of such groups include, but are not limited to, alkyl thioethers, benzyl and substituted benzyl thioethers, triphenylmethyl thioethers, and trichloroethoxycarbonyl thioester, to name but a few.

[0059] *Reference:* As used herein describes a standard or control relative to which a comparison is

performed. For example, in some embodiments, an agent, animal, individual, population, sample, sequence or value of interest is compared with a reference or control agent, animal, individual, population, sample, sequence or value. In some embodiments, a reference or control is tested and/or determined substantially simultaneously with the testing or determination of interest. In some embodiments, a reference or control is a historical reference or control, optionally embodied in a tangible medium. Typically, as would be understood by those skilled in the art, a reference or control is determined or characterized under comparable conditions or circumstances to those under assessment. Those skilled in the art will appreciate when sufficient similarities are present to justify reliance on and/or comparison to a particular possible reference or control.

[0060] *Specificity:* As is known in the art, “specificity” is a measure of the ability of a particular ligand (e.g., an agent) to distinguish its binding partner (e.g., beta-catenin) from other potential binding partners (e.g., another protein, another portion (e.g., domain) of beta-catenin).

[0061] *Subject:* As used herein, the term “subject” or “test subject” refers to any organism to which a provided compound or composition is administered in accordance with the present disclosure e.g., for experimental, diagnostic, prophylactic, and/or therapeutic purposes. Typical subjects include animals (e.g., mammals such as mice, rats, rabbits, non-human primates, and humans; insects; worms; etc.) and plants. In some embodiments, a subject may be suffering from, and/or susceptible to a disease, disorder, and/or condition. In some embodiments, a subject is a human.

[0062] *Susceptible to:* An individual who is “susceptible to” a disease, disorder, and/or condition is one who has a higher risk of developing the disease, disorder, and/or condition than does a member of the general public. In some embodiments, an individual who is susceptible to a disease, disorder and/or condition may not have been diagnosed with the disease, disorder, and/or condition. In some embodiments, an individual who is susceptible to a disease, disorder, and/or condition may exhibit symptoms of the disease, disorder, and/or condition. In some embodiments, an individual who is susceptible to a disease, disorder, and/or condition may not exhibit symptoms of the disease, disorder, and/or condition. In some embodiments, an individual who is susceptible to a disease, disorder, and/or condition will develop the disease, disorder, and/or condition. In some embodiments, an individual who is susceptible to a disease, disorder, and/or condition will not develop the disease, disorder, and/or condition.

[0063] *Therapeutic agent:* As used herein, the phrase “therapeutic agent” refers to an agent that, when administered to a subject, has a therapeutic effect and/or elicits a desired biological and/or pharmacological effect. In some embodiments, a therapeutic agent is any substance that can be used to alleviate, ameliorate, relieve, inhibit, prevent, delay onset of, reduce severity of, and/or reduce incidence of one or more symptoms or features of a disease, disorder, and/or condition.

[0064] *Therapeutic regimen:* A “therapeutic regimen”, as that term is used herein, refers to a dosing regimen whose administration across a relevant population may be correlated with a desired or beneficial therapeutic outcome.

[0065] *Therapeutically effective amount:* As used herein, the term “therapeutically effective amount”

means an amount of a substance (*e.g.*, a therapeutic agent, composition, and/or formulation) that elicits a desired biological response when administered as part of a therapeutic regimen. In some embodiments, a therapeutically effective amount of a substance is an amount that is sufficient, when administered to a subject suffering from or susceptible to a disease, disorder, and/or condition, to treat, diagnose, prevent, and/or delay the onset of the disease, disorder, and/or condition. As will be appreciated by those of ordinary skill in this art, the effective amount of a substance may vary depending on such factors as the desired biological endpoint, the substance to be delivered, the target cell or tissue, *etc.* For example, the effective amount of compound in a formulation to treat a disease, disorder, and/or condition is the amount that alleviates, ameliorates, relieves, inhibits, prevents, delays onset of, reduces severity of and/or reduces incidence of one or more symptoms or features of the disease, disorder, and/or condition. In some embodiments, a therapeutically effective amount is administered in a single dose; in some embodiments, multiple unit doses are required to deliver a therapeutically effective amount.

[0066] *Treat:* As used herein, the term “treat,” “treatment,” or “treating” refers to any method used to partially or completely alleviate, ameliorate, relieve, inhibit, prevent, delay onset of, reduce severity of, and/or reduce incidence of one or more symptoms or features of a disease, disorder, and/or condition. Treatment may be administered to a subject who does not exhibit signs of a disease, disorder, and/or condition. In some embodiments, treatment may be administered to a subject who exhibits only early signs of the disease, disorder, and/or condition, for example for the purpose of decreasing the risk of developing pathology associated with the disease, disorder, and/or condition.

[0067] *Unit dose:* The expression “unit dose” as used herein refers to an amount administered as a single dose and/or in a physically discrete unit of a pharmaceutical composition. In many embodiments, a unit dose contains a predetermined quantity of an active agent. In some embodiments, a unit dose contains an entire single dose of the agent. In some embodiments, more than one unit dose is administered to achieve a total single dose. In some embodiments, administration of multiple unit doses is required, or expected to be required, in order to achieve an intended effect. A unit dose may be, for example, a volume of liquid (*e.g.*, an acceptable carrier) containing a predetermined quantity of one or more therapeutic agents, a predetermined amount of one or more therapeutic agents in solid form, a sustained release formulation or drug delivery device containing a predetermined amount of one or more therapeutic agents, *etc.* It will be appreciated that a unit dose may be present in a formulation that includes any of a variety of components in addition to the therapeutic agent(s). For example, acceptable carriers (*e.g.*, pharmaceutically acceptable carriers), diluents, stabilizers, buffers, preservatives, *etc.*, may be included as described *infra*. It will be appreciated by those skilled in the art, in many embodiments, a total appropriate daily dosage of a particular therapeutic agent may comprise a portion, or a plurality, of unit doses, and may be decided, for example, by the attending physician within the scope of sound medical judgment. In some embodiments, the specific effective dose level for any particular subject or organism may depend upon a variety of factors including the disorder being treated and the severity of the disorder; activity of specific active compound employed; specific composition employed;

age, body weight, general health, sex and diet of the subject; time of administration, and rate of excretion of the specific active compound employed; duration of the treatment; drugs and/or additional therapies used in combination or coincidental with specific compound(s) employed, and like factors well known in the medical arts.

[0068] Unless otherwise specified, salts, such as pharmaceutically acceptable acid or base addition salts, stereoisomeric forms, and tautomeric forms, of provided compound are included.

[0069] As used herein in the present disclosure, unless otherwise clear from context, (i) the term “a” or “an” may be understood to mean “at least one”; (ii) the term “or” may be understood to mean “and/or”; (iii) the terms “comprising”, “comprise”, “including” (whether used with “not limited to” or not), and “include” (whether used with “not limited to” or not) may be understood to encompass itemized components or steps whether presented by themselves or together with one or more additional components or steps; (iv) the term “another” may be understood to mean at least an additional/second one or more; (v) the terms “about” and “approximately” may be understood to permit standard variation as would be understood by those of ordinary skill in the art; and (vi) where ranges are provided, endpoints are included.

WNT-Pathway Activating Tumors

[0070] In various cases, secreted WNT ligands have been reported to engage cell surface receptors (e.g., LRP, FZD, etc.) in multiple cell types during mammalian development, and in a restricted set of adult cell populations, to drive a signal transduction pathway that leads to stabilization of cytosolic beta-catenin protein (see, e.g., Rim, EY, Clevers H, & Nusse R. The Wnt Pathway: From Signaling Mechanisms to Synthetic Modulators. Annual Reviews in Biochemistry, 2022: 91:15.1-15.28). It has been reported that engagement of the WNT receptor complex can lead to blockade of a constitutive ‘destruction complex’ that drives proteasome-mediated degradation of beta-catenin in the absence of WNT ligand. In some cases, the ‘destruction complex’ has been reported to contain multiple proteins, including AXIN, adenomatous polyposis coli (APC), GSK3 and CK1, that cooperate to drive beta-catenin degradation. Following stabilization of cytosolic beta-catenin, it is reportedly translocated to the nucleus and activates a transcriptional program by virtue of interaction with members of the TCF transcriptional factor family (TCF1/3/4 and LEF1). The beta-catenin:TCF complex can bind to cognate deoxyribonucleic acid (DNA) sequences and drives a gene expression program involved in cell proliferation, stem cell maintenance, and epithelial-to-mesenchymal transition (EMT).

[0071] It has been reported that dysregulation of the WNT/beta-catenin pathway frequently occurs in human cancers (see, e.g., Zhong Z, Yu J, Virshup DM, Madan B. Wnts and the hallmarks of cancer. Cancer Metastasis Rev. 2020;39(3):625-45; Du P, Han, X. Report R-35. Analysis of the mutation prevalence of Wnt pathway genes and the relationship between APC/CTNNB1 mutations and Microsatellite Instability. 2023; etc.). WNT pathway mutations, including APC loss of function (LoF) mutations and CTNNB1 (beta-catenin) activating mutations, are reported to impact cancer cell proliferation, differentiation, and apoptosis (see, e.g.,

Stamos JL, Weis WI. The beta-catenin destruction complex. *Cold Spring Harb Perspect Biol.* 2013;5(1):a007898). It has been reported that APC LoF mutations can incapacitate the beta-catenin destruction complex and can lead to subsequent beta-catenin activation (see, e.g., Stamos JL, Weis WI. The beta-catenin destruction complex. *Cold Spring Harb Perspect Biol.* 2013;5(1):a007898). LoF mutations in APC, activating mutations in beta-catenin, along with other driver WNT pathway mutations, are frequently reported in common cancer types (see, e.g., Hoadley KA, Yau C, Hinoue T, et al. Cell-of-origin patterns dominate the molecular classification of 10,000 tumors from 33 types of cancer. *Cell*, 2018;173(2):291-304.E6; Gajos-Michniewicz A, Czyz M. Wnt signaling in melanoma. *Int J Mol Sci.* 2020;21(14):4852; Stewart DJ. Wnt signaling pathway in non-small cell lung cancer. *J Natl Cancer Inst.* 2014;106(1):djt356; Teeuwssen M, Fodde R. Wnt signaling in ovarian cancer stemness, EMT, and therapy resistance. *J Clin Med.* 2019;8(10):1658; etc.). Analyses of The Cancer Genome Atlas (TCGA) dataset revealed that the prevalence of these WNT pathway-activating mutations (WPAMs) is 80.7, 33.1, 21.1 and 4.5-7.2% in CRC, liver cancer, gastric cancer and non-small cell lung cancer (NSCLC), respectively (see, e.g., Du P, Han, X. Report R-35. Analysis of the mutation prevalence of Wnt pathway genes and the relationship between APC/CTNNB1 mutations and Microsatellite Instability. 2023; Hoadley KA, Yau C, Hinoue T, et al. Cell-of-origin patterns dominate the molecular classification of 10,000 tumors from 33 types of cancer. *Cell*, 2018;173(2):291-304.E6; etc.).

[0072] The functional relevance of key WPAM mutations in the initiation and maintenance of cancer has been reported preclinically. For instance, LoF mutations in the APC gene observed in patient colorectal tumor samples have been reported to drive tumorigenesis in cell and animal models of cancer (see, e.g., Bian J, Dannappel M, Wan C, et al. Transcriptional Regulation of Wnt/ β -Catenin Pathway in Colorectal Cancer. *Cells*, 2020: 9:2125). Genetic perturbation of APC gene in mouse models is reported to lead to intestinal polyps and tumorigenesis, particularly in combination with additional cancer driver mutations, like KRAS, TP53 and SMAD4 (all of which have been reported to be commonly observed in CRC patients). It has been reported that restoration of wild-type APC, or complete blockade of beta-catenin activity (using short hairpin RNA [shRNA], CRISPR) in preclinical cancer models carrying APC mutations, leads to strong inhibition of cancer cell and tumor growth, and knockdown of TCF proteins in tumor cells with activated beta-catenin also leads to inhibition of cancer cell proliferation.

[0073] It has been reported that the majority of human colorectal cancer (>80%), and many cancers representing multiple additional indications (e.g., hepatocellular, gastric, endometrial and non-small cell lung cancers), harbor mutations in genes of the WNT/beta-catenin pathway. APC and CTNNB1 (encoding β -catenin) are two of the most frequently mutated genes in this pathway. These mutations lead to the stabilization of beta-catenin at the protein level and the hyperactivation of its transcriptional activity in the nucleus of tumor cells, in complex with TCF family proteins.

[0074] Transcriptional programs driven by beta-catenin:TCF complex have been reported to activates multiple biological pathways that promote tumor growth including cell cycle regulators and the C-MYC

transcription factor, which itself is a well-described oncogenic driver (see, e.g., Rennoll S, Yochum G. 2015. Regulation of MYC gene expression by aberrant Wnt/B-catenin signaling in colorectal cancer. *World J Biol Chem.* 6(4):290-300)). Directly blocking β -catenin:TCF interaction offers a useful strategy to treat tumors carrying WPAMs. It has been an area of intense interest for decades, but few if any safe and effective therapeutics in this area have been reported and approved.

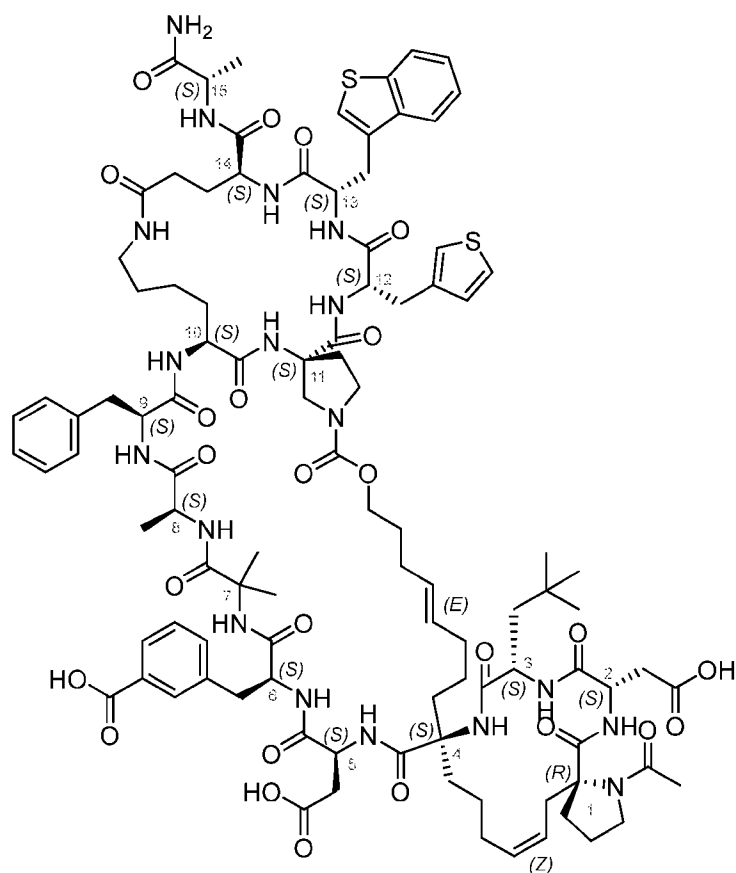
[0075] Various in vitro and in vivo assays have confirmed that I-66 can modulate beta-catenin:TCF interactions and can inhibit tumor growth including in various PDX tumor models. Among other things, the present disclosure provides technologies, e.g., formulations, doses, dosage regimens, etc., for using I-66 to treat cancers in human subjects.

I-66

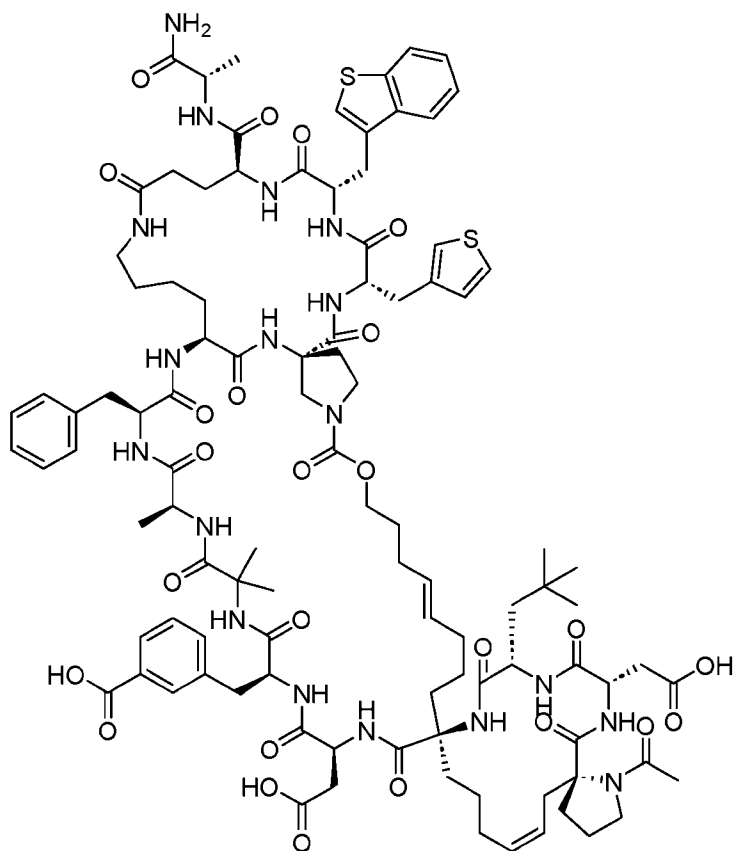
[0076] In some embodiments, the present disclosure provides technologies for manufacturing, formulating and utilizing I-66 for treating conditions, disorders or diseases including cancer.

[0077] The linear sequence of I-66 is Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOHF⁶-Aib⁷-Ala⁸-Phc⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂; it has olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹, and a lactam staple between Lys¹⁰ and Glu¹⁴. I-66 has such a structure that it has NMR spectra as shown in Figure 1, Figure 2, Figure 3, Figure 4, Figure 5, Figure 6, Figure 7, Figure 8, Figure 9, Figure 10, Figure 11 and/or Figure 12. In some embodiments, ¹³C NMR in MeOD at 25°C (e.g., 900MHz Bruker AVANCE III system with a cryoprobe, sample concentration 10 mg/750 uL) of I-66 in a preparation or composition thereof comprises or consists of the following peaks (chemical shift (δ , ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58. In some embodiments, ¹H NMR in MeOD at 25°C (e.g., 900MHz Bruker AVANCE III system with a cryoprobe, sample concentration 10 mg/750 uL) of I-66 in a preparation or composition thereof comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23. In some embodiments, ¹H NMR in MeOD at 25°C (e.g., 900MHz Bruker AVANCE III system with a cryoprobe, sample concentration 10 mg/750 uL)

of I-66 in a preparation or composition thereof comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise. In some embodiments, ^1H NMR in MeOD at 25°C (e.g., 900MHz Bruker AVANCE III system with a cryoprobe, sample concentration 10 mg/750 μL) of I-66 in a preparation or composition thereof comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69. In some embodiments, ^1H NMR in MeOD at 25°C (e.g., 900MHz Bruker AVANCE III system with a cryoprobe, sample concentration 10 mg/750 μL) of I-66 in a preparation or composition thereof comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise. As those skilled in the art readily appreciate, chemical shifts of the same compound may be deferent between or among different experiments and/or samples; despite the slight differences, those skilled in the art can determine whether two NMR results are of the same compound. Identity of I-66 is confirmed by various technologies including NMR as described herein. Various stereoisomers, including *E/Z* isomers of each double bond in the staples, may exist. I-66 is the isomer that has the characterization data, including NMR and HPLC data, that described herein. In some embodiments, I-66 is referred to by the following structure:



; in some embodiments, numbering and/or stereochemistry (e.g., one or more of (S), (R), (E), and (Z)) may be omitted, for example:



[0078] The molecular formula of I-66 is $C_{102}H_{134}N_{18}O_{25}S_2$. The molecular weight of I-66 is 2076.42.

[0079] In some embodiments, I-66 is referred to by the chemical name of 2,2'-((1''S,2R,2'S,5'S,8'S,10'E,12'Z,18''S,21''S,27''S,30''S,33''S,42''S,45''S,48''S)-1-acetyl-42'-(((S)-1-amino-1-oxopropan-2-yl)carbamoyl)-45''-(benzo[b]thiophen-3-ylmethyl)-30''-benzyl-21''-(3-carboxybenzyl)-24'',24'',27''-trimethyl-5'-neopentyl-3',5'',6',16',16'',19'',22'',25'',28'',31'',39'',44'',47'',50'',52''-pentadeca-oxo-48''-(thiophen-3-ylmethyl)-6''-oxa-1',4',4'',7',17'',20'',23'',26'',29'',32'',38'',43'',46'',49'',51''-pentadecaazadispiro[pyrrolidine-2,15'-cyclohexadecane-8',15''-tricyclo[31.17.2.11,4]tripentacontane]-10'',12'-dien-2',18''-diyl)diacetic acid.

[0080] In some embodiments, I-66 is provided as a white or slightly colored solid. In some embodiments, dissociate constant (pKa) is reported to be about 5.0 (all carboxylic acids). In some embodiments, I-66 is formulated as a salt to provide higher solubility compared to free acid. Some salts, e.g., sodium salt, of I-66 have solubility of > 100 mg/mL in water compared to < 26 mg/mL for free acid. I-66 preparations may be hygroscopic under certain conditions. For example, for a sodium salt preparation, 13% wt gain from 40 to 80% RH is reported, and max wt gain ~30% is reported. In some embodiments, I-66 preparations are amorphous. In some embodiments, an I-66 preparation is amorphous sodium salt. In some embodiments, a sodium salt is a trisodium salt. Unless noted otherwise, an I-66 sodium salt is trisodium salt in the present disclosure.

Compositions

[0081] Among other things, the present disclosure provides compositions that comprise I-66. In some embodiments, I-66 in a composition exists as a salt form. In some embodiments, I-66 in a composition exists as a pharmaceutically acceptable salt form. In some embodiments, multiple forms of I-66 may exist in a composition. In some embodiments, I-66 is provided as a trisodium salt, e.g., as a prepared drug substance.

[0082] In some embodiments, the present disclosure provides technologies for formulating I-66 to provide compositions of sufficient concentrations and stability, e.g., for clinical uses. In some embodiments, the present disclosure provides an I-66 drug product. In some embodiments, an I-66 pharmaceutical composition is an I-66 drug product. In some embodiments, a composition is a liquid composition.

[0083] In some embodiments, a composition comprises or consists of:

- I-66;
- a solubilizing agent;
- a buffer agent;
- optionally a pH adjustment agent; and
- a solvent.

[0084] In some embodiments, a composition comprises or consists of:

- I-66;
- an amino acid;
- a buffer agent;
- optionally a pH adjustment agent; and
- a solvent.

[0085] In some embodiments, a composition comprises or consists of:

- I-66;
- a PEG or a surfactant;
- a buffer agent;
- optionally a pH adjustment agent; and
- a solvent.

[0086] In some embodiments, a composition comprises or consists of:

- I-66;
- a PEG;
- a buffer agent;
- optionally a pH adjustment agent; and
- a solvent.

[0087] In some embodiments, a composition comprises or consists of:

- I-66;
- a surfactant;

a buffer agent;
optionally a pH adjustment agent; and
a solvent.

[0088] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a PEG or a surfactant;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.

[0089] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a PEG;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.

[0090] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a surfactant;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.

[0091] In some embodiments, a composition comprises or consists of:

I-66;
a solubilizing agent;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.

[0092] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.

[0093] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0094] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0095] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0096] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0097] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0098] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0099] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-20 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0100] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-20 mM;
optionally sodium hydroxide;

optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0101] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-20 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0102] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0103] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0104] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0105] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0106] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0107] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0108] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0109] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0110] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0111] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0112] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0113] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0114] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-15 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0115] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0116] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% v/v;

a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;

optionally a pH adjustment agent; and

water;

wherein the pH of the composition is about 7.0-9.0.

[0117] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

arginine, wherein the concentration of arginine is about 1-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;

a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;

optionally a pH adjustment agent; and

water;

wherein the pH of the composition is about 7.0-9.0.

[0118] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

arginine, wherein the concentration of arginine is about 1-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;

optionally a pH adjustment agent; and

water;

wherein the pH of the composition is about 7.0-9.0.

[0119] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

arginine, wherein the concentration of arginine is about 1-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid; and

water;

wherein the pH of the composition is about 7.0-9.0.

[0120] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0121] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0122] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-20 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0123] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-20 mM;

optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0124] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-20 mM;

optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0125] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;

optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0126] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;

optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0127] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;

arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

[0128] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0129] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0130] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0131] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

[0132] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0133] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;

arginine, wherein the concentration of arginine is about 5-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0134] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;

arginine, wherein the concentration of arginine is about 5-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0135] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;

arginine, wherein the concentration of arginine is about 5-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0136] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-25 mg/mL;

arginine, wherein the concentration of arginine is about 10 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0137] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 5-15 mg/mL;

arginine, wherein the concentration of arginine is about 10 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0138] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 10 mg/mL;

arginine, wherein the concentration of arginine is about 10 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and

a buffer;

wherein the pH of the composition is about 7.0-9.0.

[0139] I-66 in a composition may exist in various forms. In some embodiments, a form of I-66 is a pharmaceutically acceptable salt form. In some embodiments, I-66 is exist as one or more dissolved pharmaceutically acceptable salt form. In some embodiments, there are two or more forms of I-66 in a composition. In some embodiments, concentration of I-66 is about 1-25 mg/mL (unless otherwise noted, assessed as free acid). In some embodiments, concentration of I-66 is about 10 mg/mL. In some embodiments, concentration of I-66 is about 20 mg/mL. In some embodiments, concentration of I-66 is about 25 mg/mL.

[0140] In some embodiments, there is one solubilizing agent in a composition. In some embodiments, there are two or more solubilizing agents in a solution. In some embodiments, a solubilizing agent is an amino acid. In some embodiments, a solubilizing agent is a basic amino acid. In some embodiments, a solubilizing agent is arginine. In some embodiments, a solubilizing agent is PEG. In some embodiments, a solubilizing agent is PEG-400. In some embodiments, a solubilizing agent is a surfactant. In some embodiments, a solubilizing agent is a nonionic surfactant. In some embodiments, a solubilizing agent is Tween 80. In some embodiments, each solubilizing agent is independently selected from an amino acid, a polyethylene glycol, and a surfactant. In some embodiments, each solubilizing agent is independently selected from an amino acid, a polyethylene glycol, and a nonionic surfactant. In some embodiments, a solubilizing agent is a basic amino acid. In some embodiments, there are two solubilizing agent in a composition. In some embodiments, a composition comprises two or more solubilizing agents, for example, in some embodiments, a composition comprises both arginine and PEG-400 as solubilizing agent. In some embodiments, a solubilizing agent is Tween 80. In some embodiments, concentration of a solubilizing agent is about 1-20 mg/mL. In some embodiments, concentration of a basic amino acid is about 1-20 mg/mL. In some embodiments, concentration of arginine is about 1-20 mg/mL. In some embodiments, concentration of arginine is about 10 mg/mL. In some embodiments, concentration of a solubilizing agent, e.g., of a PEG, a surfactant, etc., is about 0.5%-5% (w/v). In some embodiments, concentration of PEG is about 0.5%-5% (w/v). In some embodiments, concentration of PEG is about 2%-5% (w/v). In some embodiments, concentration of PEG is about 3% (w/v). In some embodiments, concentration of PEG-400 is about 0.5%-5% (w/v). In some embodiments, concentration of PEG-400 is about 2%-5% (w/v). In some embodiments, concentration of PEG-400 is about 3% (w/v). In some embodiments, concentration of PEG-400 is about 3.4% (w/v). For example, as shown in Table E-8, concentration of PEG-400 is 0.336 mg/mL. In some embodiments, concentration of a surfactant is about 0.5%-5% (w/v). In some embodiments, concentration of PEG is about 2%-5% (w/v). In some embodiments, concentration of PEG is about 3% (w/v). In some embodiments, concentration of Tween 80 is about 0.01%-5%, 0.05%-2%, 0.1%-2%, 0.5%-2%, or 0.5-1.5% (w/v). In some embodiments, concentration of Tween 80 is about 1% (w/v). In some embodiments, a concentration described herein, e.g., for PEG-400, is v/v instead of w/v; in some embodiments, a composition

is otherwise identical except that the concentration is v/v instead of w/v. In some embodiments, concentration of a solubilizing agent, e.g., of a PEG, a surfactant, etc., is about 0.5%-5% (v/v). In some embodiments, concentration of PEG is about 0.5%-5% (v/v). In some embodiments, concentration of PEG is about 2%-5% (v/v). In some embodiments, concentration of PEG is about 3% (v/v). In some embodiments, concentration of PEG-400 is about 0.5%-5% (v/v). In some embodiments, concentration of PEG-400 is about 2%-5% (v/v). In some embodiments, concentration of PEG-400 is about 3% (v/v). In some embodiments, concentration of PEG-400 is about 3.4% (v/v). For example, as shown in Table E-8, concentration of PEG-400 is 0.336 mg/mL. In some embodiments, concentration of a surfactant is about 0.5%-5% (v/v). In some embodiments, concentration of PEG is about 2%-5% (v/v). In some embodiments, concentration of PEG is about 3% (v/v). In some embodiments, concentration of Tween 80 is about 0.01%-5%, 0.05%-2%, 0.1%-2%, 0.5%-2%, or 0.5-1.5% (v/v). In some embodiments, concentration of Tween 80 is about 1% (v/v).

[0141] In some embodiments, w/v may be replaced with w/w and vice versa when density is about 1.

[0142] In some embodiments, a buffering agent is a phosphate salt. In some embodiments, a buffering agent is sodium phosphate monobasic. In some embodiments, a buffering agent is Tris. In some embodiments, concentration of a buffering agent is about 10 mM.

[0143] In some embodiments, a composition is or comprises a buffer. Various pharmaceutically acceptable buffer system may be utilized in accordance with the present disclosure. For example, in some embodiments, a buffer is a phosphate buffer. In some embodiments, a buffer is a Tris buffer.

[0144] In some embodiments, a composition comprises a pharmaceutically acceptable salt. In some embodiments, a pharmaceutically acceptable salt is not a salt of I-66, a solubilizing agent, an amino acid or a buffering agent. In some embodiments, a pharmaceutically acceptable salt is NaCl. In some embodiments, a pharmaceutically acceptable salt is formed when adjusting pH (e.g., when HCl is added or when NaOH is added).

[0145] Various pH adjustment agents may be utilized in accordance with the present disclosure. In some embodiments, a pH adjustment agent is NaOH. In some embodiments, a pH adjustment agent is HCl. For example, in some embodiments, a buffer is a sodium phosphate monobasic buffer whose pH is adjusted with NaOH or HCl. Those skilled in the art appreciate that typically, pH adjustments are utilized as needed to adjust pH.

[0146] In some embodiments, a composition is or comprises a buffer. In some embodiments, a buffer is a phosphate buffer.

[0147] In some embodiments, pH of a composition is about 6-9. In some embodiments, pH of a composition is about 6.5-9.0. In some embodiments, pH of a composition is about 7.0-9.0. In some embodiments, pH of a composition is about 7.5-8.0. In some embodiments, pH of a composition is about 7.8-8.0. In some embodiments, pH of a composition is about 7.5. In some embodiments, pH of a composition is about 7.8. In some embodiments, pH of a composition is about 8.0. In some embodiments,

pH of a composition is about 7.8-8.0.

[0148] In some embodiments, a solvent is water.

[0149] In some embodiments, a pharmaceutical composition comprises or consists of I-66, arginine, PEG 400, sodium phosphate, optionally NaOH or HCl, and water.

[0150] For example, in some embodiments, an I-66 drug product is or comprises a 20 mg/mL concentration of I-66 in a 10 mg/mL arginine / 3% PEG 400 (w/v) / 10 mM sodium phosphate pH= 7.8-8.0 solution. In some embodiments, an I-66 drug product is or comprises 10 mg/mL I-66 in a 10 mg/mL arginine / 3% PEG 400 (w/v) / 10 mM sodium phosphate pH 7.8-8.0 solution. In some embodiments, an I-66 drug product is or comprises a 20 mg/mL concentration of I-66 in a 10 mg/mL arginine / 3% PEG 400 (v/v) / 10 mM sodium phosphate pH= 7.8-8.0 solution. In some embodiments, an I-66 drug product is or comprises 10 mg/mL I-66 in a 10 mg/mL arginine / 3% PEG 400 (v/v) / 10 mM sodium phosphate pH 7.8-8.0 solution. In some embodiments, a composition comprises up to 25 mg/mL I-66 in 10 mM sodium phosphate pH 8, 10 mg/mL arginine, 6% PEG 400 solution or in a 1% Tween 80/99% 10 mM PBS pH 7.4 solution.

[0151] In some embodiments, an I-66 drug product, I-66 Injection, is supplied as a 10 mg/mL liquid in Type 1 glass vials with appropriate elastomer stoppers and aluminum seals. In some embodiments, I-66 Injection contains 10 mg/mL I-66, 10 mg/mL L-arginine and 33.6 mg/mL PEG 400 as excipients, in 10 mM sodium phosphate adjusted to pH 8.0.

[0152] In some embodiments, a composition, e.g., a drug product described herein, is prepared and dosed with 5% dextrose (e.g., 5% Dextrose Injection, USP). In some embodiments, 5% dextrose is used as a control article and vehicle. In some embodiments, a drug product is diluted with a pharmaceutically acceptable diluent to a set volume for administration. In some embodiments, a pharmaceutically acceptable dilute is a dextrose solution. In some embodiments, a pharmaceutically acceptable dilute is 5% Dextrose Injection, USP. In some embodiments, a set volume is about 250 mL. In some embodiments, a drug product is diluted by about 1.1-200, 1.5-150, 1.5-100, 1.5-50, 2-100, 2-50, 1.2, 1.5, 2, 2.5, 3, 4, 5, 6, 7, 8, 9, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 100, 150, 200 fold, e.g., depending on amount to be administered.

[0153] In some embodiments, a composition comprises or consists of:

- I-66;
- a solubilizing agent;
- a buffer agent;
- optionally a pH adjustment agent;
- a sugar; and
- a solvent.

[0154] In some embodiments, a composition comprises or consists of:

- I-66;
- an amino acid;

a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0155] In some embodiments, a composition comprises or consists of:

I-66;
a PEG or a surfactant;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0156] In some embodiments, a composition comprises or consists of:

I-66;
a PEG;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0157] In some embodiments, a composition comprises or consists of:

I-66;
a surfactant;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0158] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a PEG or a surfactant;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0159] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;

a PEG;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0160] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a surfactant;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0161] In some embodiments, a composition comprises or consists of:

I-66;
a solubilizing agent;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0162] In some embodiments, a composition comprises or consists of:

I-66;
an amino acid;
a buffer agent;
optionally a pH adjustment agent;
a sugar; and
a solvent.

[0163] Useful solubilizing agents, buffer agents, amino acids, PEGs, surfactants, pH adjustment agents, sugars and solvents, e.g., are as described herein. For example, in some embodiments, a sugar is dextrose. In some embodiments, the present disclosure provides compositions described below.

[0164] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and water.

[0165] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0166] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0167] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0168] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

[0169] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

[0170] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

[0171] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water.

[0172] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0173] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0174] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0175] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0176] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

[0177] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0178] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0179] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0180] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

[0181] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

[0182] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;

optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

[0183] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;

optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water.

[0184] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0185] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0186] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0187] In some embodiments, a composition comprises or consists of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

[0188] In some embodiments, concentration of I-66 is about 0.1-6 mg/mL. In some embodiments, it is about 0.2-5 mg/mL. Certain concentrations are presented in Example 3 as examples.

[0189] In some embodiments, concentration of a solubilizing agent is about 0.1-6 mg/mL. In some embodiments, concentration of an amino acid is about 0.1-6 mg/mL. In some embodiments, concentration of arginine is about 0.1-6 mg/mL. In some embodiments, it is about 0.2-5 mg/mL. Certain concentrations are presented in Example 3 as examples.

[0190] In some embodiments, concentration of a PEG is about 0.01%-3% v/v. In some embodiments, concentration of PEG-400 is about 0.01%-3% v/v. In some embodiments, it is about 0.05%-2% v/v. In some embodiments, concentration of a PEG is about 0.01%-3% w/v. In some embodiments, concentration of PEG-400 is about 0.01%-3% w/v. In some embodiments, it is about 0.05%-2% w/v. Certain concentrations are presented in Example 3 as examples.

[0191] In some embodiments, concentration of a surfactant is about 0.01%-3% v/v. In some embodiments, it is about 0.05%-2% v/v. In some embodiments, concentration of a surfactant is about 0.01%-3% w/v. In some embodiments, it is about 0.05%-2% w/v.

[0192] In some embodiments, concentration of dextrose is about 0.01-0.05 mg/mL. In some embodiments, it is about 0.02-0.05 mg/L. Certain concentrations are presented in Example 3 as examples.

[0193] In some embodiments, concentration of a buffer or a buffer agent or sodium phosphate monobasic is about 0.01-100 mM. In some embodiments, it is about 0.01-50 mM. In some embodiments, it is about 0.01-20 mM. In some embodiments, it is about 0.01-10 mM. In some embodiments, it is about 0.1-10 mM. In some embodiments, it is about 0.2-10 mM.

[0194] In some embodiments, pH of a composition is about 7.0-9.0 as described herein.

[0195] Among other things, provided compositions provide high solubility of I-66.

[0196] In some embodiments, the present disclosure provides a method for preparing a composition, comprising dissolving various components (e.g., I-66, solubilizing agents, amino acids, arginine, surfactants,

PEG-400, buffering agents, sodium phosphate monobasic, etc.) and adjusting pH with a pH adjusting agents. In some embodiments, a component may be utilized as a salt form, e.g., a pharmaceutically acceptable salt such as a sodium salt form. In some embodiments, a form comprises water, e.g., a hydrate form. In some embodiments, a form is a hydrate of a pharmaceutically acceptable salt. In some embodiments, the present disclosure provides a method for preparing a composition for administration, e.g., IV, comprising diluting an I-66, e.g., a composition comprising a higher concentration of I-66, e.g., about 1-25 mg/mL, about 5-25 mg/mL, about 5-20 mg/mL, about 5-15 mg/mL, about 5 mg/mL, about 10 mg/mL, about 15 mg/mL, about 15 mg/mL, about 25 mg/mL, etc., with a pharmaceutically acceptable diluent as described herein (e.g., 5% Dextrose for Injection, USP). In some embodiments, the composition for administration provides the amount of I-66 to be administered to a patient as described herein. In some embodiments, a volume of composition for administration is about 250 mL.

[0197] Various compositions and methods are presented in the Examples as examples.

[0198] In some embodiments, a composition, e.g., a liquid I-66 composition is stored at 2-8 °C. In some embodiments, a preparation of I-66, e.g., a drug substance, is stored at -20 °C

[0199] Multiple stereoisomers exist for I-66. In some embodiments, level of I-66 in a composition is enriched relative to one or more or all of its stereoisomers. For example, in some embodiments, a particular configuration of a double bond (*E/Z*) is enriched. In some embodiments, for each double bond a configuration is independently enriched. In some embodiments, for a chiral element, e.g., a chiral center, one configuration is enriched. In some embodiments, for a chiral center bonded to two staples, one configuration is enriched. In some embodiments, for each chiral element a configuration is independently enriched. In some embodiments, for one or more or all stereochemical element (e.g., double bonds, chiral element, etc.), one configuration is independently enriched. In some embodiments, for each double bond in each staple, one configuration is independently enriched. In some embodiments, for each double bond in each staple, one configuration is independently enriched, and for a chiral center bonded to two staples, one configuration is enriched. In some embodiments, enrichment for each double bond is independently *E* or *Z*. In some embodiments, enrichment for each chiral element is independently *R* or *S*. In some embodiments, enrichment for each stereochemical element, e.g., double bond, chiral center, etc., is about or at least about a certain level, e.g., 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% (percentage of an agent). In some embodiments, about or at least about a certain level, e.g., 85%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, or 99% of all molecules in a composition that share the constitution of an agent or a salt thereof are the agent or a salt thereof. In some embodiments, it is about or at least about 85%. In some embodiments, it is about or at least about 90%. In some embodiments, it is about or at least about 95%. In some embodiments, it is about or at least about 96%. In some embodiments, it is about or at least about 97%. In some embodiments, it is about or at least about 98%. In some embodiments, it is about or at least about 99%.

[0200] In some embodiments, purity of I-66 is about or at least about 85%, 90%, 91%, 92%, 93%, 94%,

95%, 96%, 97%, 98%, or 99%. In some embodiments, it is about or at least about 85%. In some embodiments, it is about or at least about 90%. In some embodiments, it is about or at least about 95%. In some embodiments, it is about or at least about 96%. In some embodiments, it is about or at least about 97%. In some embodiments, it is about or at least about 98%. In some embodiments, it is about or at least about 99%.

[0201] In some embodiments, the present disclosure provides a method, comprising comparing I-66 in a preparation with a reference standard. In some embodiments, a method is utilized for confirming identify of I-66 in a preparation. In some embodiments, a method is utilized for quality control of an I-66 preparation. In some embodiments, a method is utilized for releasing an I-66 preparation (e.g., purify of I-66 above a certain level and/or impurities are below certain individual and/or total levels). In some embodiments, a reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ¹H NMR in MeOD at 25°C, its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift

(δ , ppm): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

[0202] Those skilled in the art reading the present disclosure appreciate that various technologies including analytical chemistry technologies may be utilized in accordance with the present disclosure. For example, in some embodiments, HPLC is utilized. Various useful HPLC technologies are utilized here as examples. In some embodiments, HPLC condition is as described in Example 1. In some embodiments, HPLC condition is as described in Table E-7. In some embodiments, HPLC condition is as described in Table E-10. In some embodiments, NMR is utilized. In some embodiments, NMR is or comprises ^1H NMR. In some embodiments, NMR is or comprises ^{13}C NMR. In some embodiments, NMR is or comprises ^{15}N NMR. In some embodiments, NMR is or comprises NOE. In some embodiments, NMR is or comprises two-dimensional NMR. In some embodiments, NMR is or comprises multi-dimensional NMR. In some embodiments, each dimension independently comprises or is ^1H or ^{13}C NMR. In some embodiments, each dimension independently comprises or is ^1H , ^{13}C or ^{15}N NMR.

Uses and Applications

[0203] Provided technologies, e.g., I-66, its preparations and compositions can be utilized for various purposes.

[0204] In some embodiments, the present disclosure provides a method for treating cancer, comprising administering or delivering to a human subject suffering therefrom an effective amount of I-66. In some embodiments, the present disclosure provides a method for treating cancer, comprising administering or to a human subject suffering therefrom a pharmaceutical composition as described herein. In some embodiments, a cancer is colorectal cancer. In some embodiments, a subject has a solid tumor. In some embodiments, a subject has advanced and/or metastatic solid tumor. In some embodiments, a subject has locally advanced tumors. In some embodiments, a subject has locally advanced solid tumors. In some embodiments, a subject has metastatic cancer. In some embodiments, a subject has metastatic solid tumors. In some embodiments, a

subject has locally advanced and metastatic solid tumors. In some embodiments, a cancer is gastric cancer. In some embodiments, a cancer is lung cancer. In some embodiments, a lung cancer is nonsquamous. In some embodiments, a lung cancer is squamous. In some embodiments, a cancer is non-small cell lung cancer. In some embodiments, a cancer is non-small cell carcinoma. In some embodiments, a cancer is non-small cell lung cancer metastatic. In some embodiments, a cancer is non-small cell lung cancer stage IIIB. In some embodiments, a cancer is non-small cell carcinoma of lung, TNM stage 4. In some embodiments, a cancer is gastroesophageal-junction cancer. In some embodiments, a subject has solid tumors with a Wnt-pathway activating mutation (WPAM). In some embodiments, a subject has microsatellite stable (MSS) colorectal cancer. In some embodiments, a cancer is non-small cell lung cancer with a Wnt-pathway activating mutation (WPAM) in APC or beta-catenin. In some embodiments, a cancer is non-small cell lung cancer with WPAM in APC. In some embodiments, a cancer is non-small cell lung cancer with WPAM in beta-catenin. In some embodiments, a cancer is gastric cancer/gastroesophageal junction carcinoma (GEJ) with a Wnt-pathway activating mutation (WPAM) in APC or beta-catenin. In some embodiments, a cancer is gastric cancer with WPAM in APC. In some embodiments, a cancer is gastric cancer with WPAM in beta-catenin. In some embodiments, a cancer is gastroesophageal junction carcinoma with WPAM in APC. In some embodiments, a cancer is gastroesophageal junction carcinoma with WPAM in beta-catenin. In some embodiments, a cancer is refractory gastric cancer/gastroesophageal junction carcinoma (GEJ) with a Wnt-pathway activating mutation (WPAM) in APC or beta-catenin. In some embodiments, a cancer is refractory gastric cancer with WPAM in APC. In some embodiments, a cancer is refractory gastric cancer with WPAM in beta-catenin. In some embodiments, a cancer is refractory gastroesophageal junction carcinoma with WPAM in APC. In some embodiments, a cancer is refractory gastroesophageal junction carcinoma with WPAM in beta-catenin. In some embodiments, a subject has solid tumors with a Wnt-pathway activating mutation. In some embodiments, a subject has solid tumors with WPAM in APC. In some embodiments, a subject has solid tumors with WPAM in beta-catenin. In some embodiments, a solid tumor is non-MSI-H. In some embodiments, a solid tumor is non-dMMR. In some embodiments, a subject has advanced or metastatic solid tumor that is non-MSI-H or non-dMMR CRC. In some embodiments, a cancer is advanced or metastatic NSCLC with a WPAM mutation, e.g., in APC or beta-catenin. In some embodiments, a cancer is refractory advanced or metastatic NSCLC with a WPAM mutation, e.g., in APC or beta-catenin. In some embodiments, NSCLC is nonsquamous. In some embodiments, NSCLC is squamous. In some embodiments, a cancer is advanced or metastatic gastric with a WPAM mutation, e.g., in APC or beta-catenin. In some embodiments, a cancer is refractory advanced or metastatic gastric cancer with a WPAM mutation, e.g., in APC or beta-catenin. In some embodiments, a cancer is advanced or metastatic GEJ with a WPAM mutation, e.g., in APC or beta-catenin. In some embodiments, a cancer is refractory advanced or metastatic GEJ with a WPAM mutation, e.g., in APC or beta-catenin.

[0205] In some embodiments, a cancer is refractory. In some embodiments, a subject has refractory advanced or metastatic solid tumor with a WPAM mutation.

[0206] In some embodiments, a subject has WNT pathway activating mutation. In some embodiments, a tumor has WNT pathway activating mutation. In some embodiments, a tumor is microsatellite stable.

[0207] In some embodiments, Wnt pathway activating mutation is a mutation in APC. In some embodiments, Wnt pathway activating mutation is a mutation in beta-catenin. In some embodiments, a Wnt pathway activating mutation is selected from APC loss of function mutations (LOF, including frameshift, nonsense, or splice site), *CTNNB1* gain of function mutations (GOF, including missense, in-frame insertion, or deletion at codons 32-37, 41, and 45), *RNF43* LOF mutations, and *RSPO2* and *RSPO3* fusions. In some embodiments, a Wnt pathway activating mutation is selected from mutations in APC, *CTNNB1*, *RNF43*, *RSPO2* and *RSPO3*. In some embodiments, a Wnt pathway activating mutation is selected from mutations in other Wnt-pathway genes, such as *AMER1*, *AXIN1*, *AXIN2*, *BCL9*, *CSNK1A1*, *GSK3B*, *LRP5*, *LRP6*, *LGR5*, *TCF7L2*, and *WIF1*.

[0208] In some embodiments, I-66 is administered or delivered intravenously. In some embodiments, I-66 is provided as a sterile solution for intravenous (IV) administration. In some embodiments, I-66 injection, 10 mg/mL, is supplied as a sterile, aseptically processed solution labeled for storage at 2-8°C in single-use vials for intravenous (IV) administration. In some embodiments, an I-66 injection is diluted using a pharmaceutically acceptable diluent, e.g., 5% Dextrose Injection, USP, to a set volume, e.g., 250 mL for administration, e.g., intravenous administration. Various I-66 compositions for delivering or administering I-66 are as described herein. As described herein, I-66 can be delivered or administered as various forms including pharmaceutically acceptable salt such as sodium salt forms. In some embodiments, I-66 is delivered as a trisodium salt. In some embodiments, I-66 is administered as a trisodium salt. In some embodiments, I-66 in a composition is in a trisodium salt form. In some embodiments, I-66 in a composition is dissolved trisodium salt.

[0209] In some embodiments, a dosing schedule is weekly. In some embodiments, a dosing schedule is weekly in 28-day cycles. In some embodiments, a dosing schedule is weekly in 28-day cycles. In some embodiments, I-66 is administered or delivered about once weekly in continuous cycles of about 21 or about 28 days. In some embodiments, I-66 is administered or delivered intravenously about once weekly in continuous cycles of about 21 or about 28 days. In some embodiments, each dose in a cycle contains or delivers about the same amount of I-66. In some embodiments, doses of several (e.g., 2, 3, 4, 5 or more) or all cycles contain or deliver about the same amount of I-66. In some embodiments, all doses for a subject are about the same. In some embodiments, a dose is different (higher or lower) than another dose. In some embodiments, dose of a cycle is different (higher or lower) than dose of another cycle. In some embodiments, a cycle is about 21 days. In some embodiments, each cycle is independently about 21 days. In some embodiments, a cycle is about 28 days. In some embodiments, each cycle is independently about 28 days. In some embodiments, a dose is about 18 mg/m² (unless noted otherwise, of I-66 free acid). In some embodiments, a dose is about 36 mg/m². In some embodiments, a dose is about 72 mg/m². In some embodiments, a dose is about 144 mg/m². In some embodiments, a dose is about 240 mg/m². In some

embodiments, a dose is about 360 mg/m². In some embodiments, a dose is about 480 mg/m². In some embodiments, a dose is about 600 mg/m². In some embodiments, a dose of a cycle is about 18 mg/m². In some embodiments, a dose of a cycle is about 36 mg/m². In some embodiments, a dose of a cycle is about 72 mg/m². In some embodiments, a dose of a cycle is about 144 mg/m². In some embodiments, a dose of a cycle is about 240 mg/m². In some embodiments, a dose of a cycle is about 360 mg/m². In some embodiments, a dose of a cycle is about 480 mg/m². In some embodiments, a dose of a cycle is about 600 mg/m². Certain useful technologies for calculating body surface area and amounts of I-66 are described in the Examples. As appreciated by those skilled in the art, in a pharmaceutical composition such as a drug product, I-66 may exist in one or more forms including one or more pharmaceutically acceptable salt forms. For example, in some embodiments, I-66 may exist as one or more dissolved salt forms (e.g., sodium salt). In some embodiments, a patient may start with a lower dose. In some embodiments, a patient may start with a higher dose. In some embodiments, there are one or more loading doses. In some embodiments, there are no loading doses.

[0210] In some embodiments, I-66 is administered or delivered about every two weeks. In some embodiments, I-66 is administered or delivered about every three weeks.

[0211] In some embodiments, after one or more doses effects of I-66 are assessed. In some embodiments, biopsies may be taken from a subject to assess one or more biomarkers, e.g., transcripts or polypeptides levels (e.g., of cMyc, Axin2, etc.). In some embodiments, assessment is compared to baseline.

[0212] In some embodiments, the present disclosure provides I-66 compositions for modulating beta-catenin functions. In some embodiments, the present disclosure provides I-66 compositions for modulating beta-catenin : TCF interactions. In some embodiments, the present disclosure provides I-66 compositions for modulating functions associated with beta-catenin : TCF interactions.

[0213] In some embodiments, provided technologies can decrease, suppress or block one or more of such interactions. In some embodiments, the present disclosure provides methods for modulating an interaction between beta-catenin and its binding partner (e.g., a TCF/LEF family member) comprising contacting beta-catenin with I-66.

[0214] In some embodiments, binding of I-66 to beta-catenin competes or inhibits binding of another agent. In some embodiments, binding of I-66 to beta-catenin competes or inhibits binding of another agent. In some embodiments, binding of I-66 to beta-catenin competes or inhibits binding of TCF or a fragment thereof.

[0215] In some embodiments, provided technologies can reduce or block beta-catenin's interactions with all TCF family members, E-cadherin and APC, but did not significantly affect its interactions with ICAT, AXIN and BCL9. In some embodiments, provided technologies can interrupt beta-catenin/TCF interaction at both physical interaction level (e.g., as confirmed by NanoBRET, co-IP, etc.) and transcriptional level (e.g., as confirmed by reporter cell line, endogenous gene expression, etc.). In some embodiments, provided technologies show no effect on beta-catenin stability.

[0216] Among other things, the present disclosure provides technologies for modulating levels and/or activities of transcripts and/or products thereof. In some embodiments, levels and/or activities of transcripts and levels and/or activities of products thereof, e.g., polypeptides, may be modulated independently, or may be increased, decreased or maintained at the same or comparable levels independently.

[0217] In some embodiments, a reduction is about or at least about 10%, 15%, 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, or 95%. In some embodiments, an increase is about or at least about 10%, 15%, 20%, 25%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, or 95%. In some embodiments, the present disclosure provide increases and/or decreases at least several days, e.g., 1, 2, 3, 4, 5, 6, 7, 8, 9, or 10 days or more after administration is completed.

[0218] In some embodiments, a system, cell, tissue, organ or subject comprises or expresses a mutant beta-catenin. In some embodiments, a system, cell, tissue, organ or subject comprises beta-catenin hyperactivity. In some embodiments, a system, cell, tissue, organ or subject comprises upregulated beta-catenin transcript level. In some embodiments, a system, cell, tissue, organ or subject comprises upregulated beta-catenin polypeptide level. In some embodiments, a system, cell, tissue, organ or subject comprises or expresses a mutant c-Myc. In some embodiments, a system, cell, tissue, organ or subject comprises c-Myc hyperactivity. In some embodiments, a system, cell, tissue, organ or subject comprises upregulated c-Myc transcript level. In some embodiments, a system, cell, tissue, organ or subject comprises upregulated c-Myc polypeptide level. In some embodiments, a system, cell, tissue, organ or subject comprises or expresses a mutant N-Myc. In some embodiments, a system, cell, tissue, organ or subject comprises N-Myc hyperactivity. In some embodiments, a system, cell, tissue, organ or subject comprises upregulated N-Myc transcript level. In some embodiments, a system, cell, tissue, organ or subject comprises upregulated N-Myc polypeptide level.

[0219] In some embodiments, a provided agent is utilized in combination with another therapy. In some embodiments, a provided agent is utilized in combination with another therapeutic agent. In some embodiments, another therapy or therapeutic agent is administered prior to an administration or delivery of a provided agent. In some embodiments, another therapy or therapeutic agent is administered at about the same time as an administration or delivery of a provided agent. In some embodiments, a provided agent and another agent is in the same pharmaceutical composition. In some embodiments, another therapy or therapeutic agent is administered subsequently to an administration or delivery of a provided agent. In some embodiments, a subject is exposed to both a provided agent and another therapeutic agent. In some embodiments, both a provided agent and another agent can be detected in a subject. In some embodiments, a provided agent is administered before another agent is cleared out by a subject or vice versa. In some embodiments, a provided agent is administered within the half-life, or 2, 3, 4, 5 or 6 times of the half-life, of another agent or vice versa. In some embodiments, a subject is exposed to a therapeutic effect of a provided agent and a therapeutic effect of another therapeutic agent. In some embodiments, an agent may provide an effect after an agent is cleared out or metabolized by a subject. In some embodiments, a procedure, e.g.,

surgery, radiation, etc., may provide an effect after the procedure is completed.

[0220] In some embodiments, another therapy is a cancer therapy. In some embodiments, another therapy is or comprises surgery. In some embodiments, another therapy is or comprises radiation therapy. In some embodiments, another therapy is or comprises immunotherapy. In some embodiments, another therapeutic agent is or comprises a drug. In some embodiments, another therapeutic agent is or comprises a cancer drug. In some embodiments, another therapeutic agent is or comprises a chemotherapeutic agent. In some embodiments, another therapeutic agent is or comprises a hormone therapy agent. In some embodiments, another therapeutic agent is or comprises a kinase inhibitor. In some embodiments, another therapeutic agent is or comprises a checkpoint inhibitor (e.g., antibodies against PD-1, PD-L1, CTLA-4, etc.). In some embodiments, a provided agent can be administered with lower unit dose and/or total dose compared to being used alone. In some embodiments, another agent can be administered with lower unit dose and/or total dose compared to being used alone. In some embodiments, one or more side effects associated with administration of a provided agent and/or another therapy or therapeutic agent are reduced. In some embodiments, a combination therapy provides improved results, e.g., when compared to each agent utilized individually. In some embodiments, a combination therapy achieves one or more better results, e.g., when compared to each agent utilized individually.

[0221] In some embodiments, another agent is a checkpoint inhibitor, an EGFR inhibitor, a VEGF inhibitor, a VEGFR inhibitor, a kinase inhibitor, or an anti-cancer drug.

[0222] In some embodiments, an additional agent is a checkpoint inhibitor. In some embodiments, an additional agent is an immune oncology agent. In some embodiments, an additional agent is an antibody against a checkpoint molecules. In some embodiments, an additional agent is an antibody of PD1, PDL-1, CTLA4, A2AR, B7-H3, B7-H4, BTLA, IDO, KIR, LAG3, TIM-s, C10orf54, *etc.* In some embodiments, an antibody is an anti-PD1 antibody. In some embodiments, an antibody is an anti-PD-L1 antibody. In some embodiments, an antibody is an anti-CTLA4.

[0223] In some embodiments, another agent is an EGFR inhibitor, *e.g.*, erlotinib, gefitinib, lapatinib, panitumumab, vandetanib, cetuximab, *etc.* In some embodiments, another agent is an VEGF and/or VEGFR inhibitor, *e.g.*, pazopanib, bevacizumab, sorafenib, sunitinib, axitinib, ponatinib, regorafenib, vandetanib, cabozantinib, ramucirumab, lenvatinib, ziv-aflibercept, *etc.* In some embodiments, another agent is a kinase inhibitor. In some embodiments, another therapeutic agent is a chemotherapeutic agent. In some embodiments, another therapeutic agent is an anti-cancer drug, *e.g.*, cyclophosphamide, methotrexate, 5-fluorouracil (5-FU), doxorubicin, mustine, vincristine, procarbazine, prednisolone, dacarbazine, bleomycin, etoposide, cisplatin, epirubicin, capecitabine, folinic acid, actinomycin, all-trans retinoic acid, azacitidine, azathioprine, bortezomib, carboplatin, chlorambucil, cytarabine, daunorubicin, docetaxel, doxifluridine, fluorouracil, gemcitabine, hydroxyurea, idarubicin, imatinib, irinotecan, mechlorethamine, mercaptopurine, mitoxantrone, paclitaxel, pemetrexed, teniposide, tioguanine, topotecan, valrubicin, vinblastine, vindesine, vinorelbine, oxaliplatin, *etc.*

[0224] Among other things, the present disclosure provides the following Embodiments:

1. A composition, comprising:
I-66;
a PEG;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
2. A composition, comprising:
I-66;
a surfactant;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
3. A composition, comprising:
I-66;
a solubilizing agent;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
4. The composition of Embodiment 3, wherein a solubilizing agent is an amino acid.
5. A composition, comprising:
I-66;
an amino acid;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
6. The composition of any one of Embodiments 3-5, further comprising a PEG.
7. The composition of any one of Embodiments 3-6, further comprising a surfactant.
8. The composition of any one of Embodiments 3-5, further comprising another solubilizing agent.
9. The composition of Embodiment 8, wherein the another solubilizing agent is a PEG.
10. The composition of Embodiment 8, wherein the another solubilizing agent is surfactant.
11. A composition, comprising:
I-66;
an amino acid;
a PEG or a surfactant;
a buffer agent;

- optionally a pH adjustment agent; and
a solvent.
12. A composition, comprising:
I-66;
an amino acid;
a PEG;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
13. A composition, comprising:
I-66;
an amino acid;
a surfactant;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
14. The composition of any one of the preceding Embodiments, wherein the composition is a liquid composition.
15. The composition of any one of the preceding Embodiments, comprising an I-66 pharmaceutically acceptable salt form.
16. The composition of any one of the preceding Embodiments, comprising an I-66 sodium salt form.
17. The composition of any one of Embodiments 1-16, wherein I-66 is Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and the ¹³C NMR of I-66 in MeOD at 25°C, comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58, and I-66 has a purity of about or at least about 90%.
18. The composition of any one of Embodiments 1-17, wherein ¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21,

7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23.

19. The composition of any one of Embodiments 1-18, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise.

20. The composition of any one of Embodiments 1-19, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69.

21. The composition of any one of Embodiments 1-20, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

22. The composition of any one of the preceding Embodiments, wherein identity of I-66 is confirmed by comparison with a reference standard.

23. The composition of Embodiment 22, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of $\text{Ac-PL3}^1\text{-Asp}^2\text{-Npg}^3\text{-B5}^4\text{-Asp}^5\text{-3COOH}^6\text{-Aib}^7\text{-Ala}^8\text{-Phe}^9\text{-Lys}^{10}\text{-PyrS2}^{11}\text{-3Thi}^{12}\text{-BztA}^{13}\text{-Glu}^{14}\text{-Ala}^{15}\text{-NH}_2$, wherein there are olefin staples between PL3^1 and B5^4 and between B5^4 and PyrS2^{11} and a lactam staple between Lys^{10} and Glu^{14} , and:

when characterized by ^{13}C NMR in MeOD at 25°C, its ^{13}C NMR comprises the following peaks (chemical shift (δ , ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ^1H NMR in MeOD at 25°C, its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

24. The composition of Embodiment 22 or 23, wherein the comparison is or comprising utilizing HPLC.
25. The composition of Embodiment 24, wherein the HPLC condition is selected from those in Example 1.
26. The composition of Embodiment 24, wherein the HPLC condition is as described in Table E-7.
27. The composition of Embodiment 24, wherein the HPLC condition is as described in Table E-10.
28. The composition of any one of Embodiments 22-27, wherein the comparison is or comprising utilizing NMR.
29. The composition of any one of Embodiments 22-28, wherein the comparison is or comprising utilizing ^1H NMR.
30. The composition of any one of Embodiments 22-29, wherein the comparison is or comprising utilizing ^{13}C NMR.
31. The composition of any one of Embodiments 22-30, wherein the comparison is or comprising utilizing two- or multi-dimensional NMR.
32. The composition of any one of the preceding Embodiments, wherein the concentration of I-66 is about 1-25 mg/mL.
33. The composition of any one of the preceding Embodiments, wherein the concentration of I-66 is about 10 mg/mL.
34. The composition of any one of the preceding Embodiments, wherein the amino acid is a basic amino acid.
35. The composition of any one of Embodiments 4-34, wherein the amino acid is arginine.
36. The composition of any one of the preceding Embodiments, wherein the concentration of the amino acid is about 1-20 mg/mL.
37. The composition of any one of the preceding Embodiments, wherein the concentration of the amino acid is about 10 mg/mL.
38. The composition of any one of the preceding Embodiments, comprising a PEG-400.
39. The composition of any one of the preceding Embodiments, wherein the concentration of the PEG is about 1%-5% w/v.
40. The composition of Embodiment 39, wherein the concentration of the PEG is about 3.4% w/v.
41. The composition of Embodiment 39, wherein the concentration of the PEG is about 3% w/v.
42. The composition of any one of Embodiments 1-38, wherein the concentration of the PEG is about 1%-5% v/v.
43. The composition of Embodiment 39, wherein the concentration of the PEG is about 3.4% v/v.
44. The composition of Embodiment 39, wherein the concentration of the PEG is about 3% v/v.
45. The composition of any one of the preceding Embodiments, comprising a surfactant.
46. The composition of any one of the preceding Embodiments, comprising a nonionic surfactant.
47. The composition of any one of the preceding Embodiments, comprising Tween 80.

48. The composition of any one of the preceding Embodiments, wherein the concentration of the surfactant is about 0.5%-5% w/v.
49. The composition of any one of the preceding Embodiments, wherein the concentration of the surfactant is about 1% w/v.
50. The composition of any one of Embodiments 1-47, wherein the concentration of the surfactant is about 0.5%-5% v/v.
51. The composition of Embodiment 50, wherein the concentration of the surfactant is about 1% v/v.
52. The composition of any one of the preceding Embodiments, wherein the buffering agent is a phosphate salt.
53. The composition of any one of the preceding Embodiments, wherein the buffering agent is sodium phosphate monobasic.
54. The composition of any one of Embodiments 1-49, wherein the buffering agent is Tris.
55. The composition of any one of the preceding Embodiments, wherein the concentration of the buffering agent is about 10 mM.
56. The composition of any one of the preceding Embodiments, comprising a pH adjusting agent.
57. The composition of Embodiment 56, wherein the pH adjusting agent is NaOH.
58. The composition of Embodiment 56, wherein the pH adjusting agent is HCl.
59. The composition of any one of the preceding Embodiments, wherein the solvent is water.
60. The composition of any one of the preceding Embodiments, comprising I-66, arginine, PEG-400, sodium phosphate buffer, and water.
61. The composition of any one of the preceding Embodiments, wherein the pH of the composition is about 7.0-9.0.
62. The composition of any one of the preceding Embodiments, comprising I-66, arginine, PEG-400, sodium phosphate, and water, and pH of about 7.0-9.0 adjusted using NaOH and HCl.
63. The composition of any one of the preceding Embodiments, consisting of I-66, arginine, PEG-400, sodium phosphate, and water, and pH of about 7.0-9.0 adjusted using NaOH and HCl.
64. The composition of any one of the preceding Embodiments, wherein the pH of the composition is about 8.0.
65. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0.

66. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0.
67. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0.
68. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
69. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;

- wherein the pH of the composition is about 7.0-9.0.
70. A composition, consisting of:
- I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - arginine, wherein the concentration of arginine is about 5-15 mg/mL;
 - PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;
 - sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
 - optionally sodium hydroxide;
 - optionally hydrochloric acid; and
 - water;
- wherein the pH of the composition is about 7.0-9.0.
71. The composition of any one of Embodiments 65-70, wherein the concentration of PEG-400 is about 3% w/v.
72. A composition, comprising:
- I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
 - a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% v/v;
 - a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
 - optionally a pH adjustment agent; and
 - water;
- wherein the pH of the composition is about 7.0-9.0.
73. A composition, comprising:
- I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - arginine, wherein the concentration of arginine is about 1-20 mg/mL;
 - PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;
 - a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
 - optionally a pH adjustment agent; and
 - water;
- wherein the pH of the composition is about 7.0-9.0.
74. A composition, comprising:
- I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - arginine, wherein the concentration of arginine is about 1-20 mg/mL;
 - PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;
 - sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
 - optionally a pH adjustment agent; and

- water;
wherein the pH of the composition is about 7.0-9.0.
75. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
76. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
77. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
78. The composition of any one of Embodiments 65-77, wherein the concentration of I-66 is about or at least about 10 mg/mL.
79. The composition of any one of Embodiments 65-77, wherein the concentration of I-66 is about 10-25 mg/mL.

80. The composition of any one of Embodiments 65-77, wherein the concentration of I-66 is about 10 mg/mL.
81. The composition of any one of Embodiments 65-80, wherein the concentration of the amino acid or arginine is about 10 mg/mL.
82. The composition of any one of Embodiments 72-81, wherein the concentration of PEG-400 is about 3% v/v.
83. The composition of any one of Embodiments 65-82, wherein the concentration of the buffer agent or sodium phosphate monobasic is about 5-20 mM.
84. The composition of any one of Embodiments 65-82, wherein the concentration of the buffer agent or sodium phosphate monobasic is about 10 mM.
85. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
86. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
87. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;

- optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
88. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
89. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
90. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
91. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

92. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
93. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
94. A composition, comprising:
I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
95. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
96. The composition of any one of Embodiments 90-95, wherein the concentration of I-66 is about or at least about 10 mg/mL.
97. The composition of any one of Embodiments 90-95, wherein the concentration of I-66 is about 10-25 mg/mL.
98. The composition of any one of Embodiments 90-95, wherein the concentration of I-66 is about 10 mg/mL.
99. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

100. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
101. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
102. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
103. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
104. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
105. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.

106. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
107. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
108. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
109. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

110. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
111. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.
112. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
113. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
114. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and

- a buffer;
wherein the pH of the composition is about 7.0-9.0.
115. A composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
116. A composition, comprising:
I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
117. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
118. The composition of any one of Embodiments 90-95, wherein the concentration of I-66 is about or at least about 10 mg/mL.
119. The composition of any one of Embodiments 90-95, wherein the concentration of I-66 is about 10-25 mg/mL.
120. The composition of any one of Embodiments 90-95, wherein the concentration of I-66 is about 10 mg/mL.
121. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
122. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and

- a buffer;
wherein the pH of the composition is about 7.0-9.0.
123. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
124. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
125. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
126. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
127. A composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0.
128. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and

- a buffer;
wherein the pH of the composition is about 7.0-9.0.
129. The composition of any one of Embodiments 90-128, wherein the buffer is a phosphate buffer.
130. The composition of any one of Embodiments 90-128, wherein the buffer is a sodium phosphate buffer.
131. The composition of any one of Embodiments 90-128, wherein the buffer is a Tris buffer.
132. The composition of any one of Embodiments 90-131, wherein the concentration of the buffer is about 5-100 mM.
133. The composition of any one of Embodiments 90-131, wherein the concentration of the buffer is about 10 mM.
134. A composition, comprising:
10 mg/mL I-66;
10 mg/mL arginine;
33.6 mg/mL PEG-400; and
10 mM sodium phosphate;
wherein the pH of the composition is about 8.0.
135. A composition, consisting of:
10 mg/mL I-66;
10 mg/mL arginine;
33.6 mg/mL PEG-400; and
10 mM sodium phosphate;
wherein the pH of the composition is about 8.0.
136. The composition of any one of Embodiments 65-135, wherein I-66 is Ac-PL³¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL³¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and the ¹³C NMR of I-66 in MeOD at 25°C, comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58, and I-66 has a purity of about or at least about 90%.
137. The composition of any one of Embodiments 65-136, wherein ¹H NMR of I-66 in MeOD at 25°C

comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23.

138. The composition of any one of Embodiments 65-137, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise.

139. The composition of any one of Embodiments 65-138, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69.

140. The composition of any one of Embodiments 65-139, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

141. The composition of any one of Embodiments 65-140, wherein identity of I-66 is confirmed by comparison with a reference standard.

142. The composition of Embodiment 141, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹

and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ¹H NMR in MeOD at 25°C, its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39,

8.15, 8.86, and 7.69, wherein each peak represents ^1H unless indicated otherwise.

143. The composition of Embodiment 141 or 142, wherein the comparison is or comprising utilizing HPLC.

144. The composition of Embodiment 143, wherein the HPLC condition is selected from those in Example 1.

145. The composition of Embodiment 143, wherein the HPLC condition is as described in Table E-7.

146. The composition of Embodiment 143, wherein the HPLC condition is as described in Table E-10.

147. The composition of any one of Embodiments 141-146, wherein the comparison is or comprising utilizing NMR.

148. The composition of any one of Embodiments 141-147, wherein the comparison is or comprising utilizing ^1H NMR.

149. The composition of any one of Embodiments 141-148, wherein the comparison is or comprising utilizing ^{13}C NMR.

150. The composition of any one of Embodiments 141-149, wherein the comparison is or comprising utilizing two- or multi-dimensional NMR.

151. The composition of any one of the proceeding Embodiment, further comprising a pharmaceutically acceptable carrier.

152. The composition of any one of the preceding Embodiments, further comprising a sugar.

153. The composition of Embodiment 152, wherein the sugar is dextrose.

154. A composition, prepared by diluting a composition of any one of Embodiments 1-153 with a pharmaceutically acceptable diluent to a set volume.

155. The composition of Embodiment 154, wherein the pharmaceutically acceptable diluent is a sugar solution.

156. The composition of Embodiment 154, wherein the pharmaceutically acceptable diluent is a dextrose solution.

157. The composition of Embodiment 154, wherein the pharmaceutically acceptable diluent is 5% Dextrose Injection, USP.

158. The composition of any one of Embodiments 154-157, wherein the set volume is 250 mL.

159. The composition of any of Embodiments 154-158, wherein the concentration of I-66 is about 0.05-10 mg/mL.

160. The composition of any of Embodiments 154-158, wherein the concentration of I-66 is about 0.05-6 mg/mL.

161. The composition of any of Embodiments 154-158, wherein the concentration of I-66 is about 0.1-5 mg/mL.

162. The composition of any one of Embodiments 154-161, wherein the concentration of the solubilizing agent, amino acid, or arginine is about 0.05-10 mg/mL.

163. The composition of any one of Embodiments 154-161, wherein the concentration of the solubilizing agent, amino acid, or arginine is about 0.05-6 mg/mL.
164. The composition of any one of Embodiments 154-161, wherein the concentration of the solubilizing agent, amino acid, or arginine is about 0.01-5 mg/mL.
165. The composition of any one of Embodiments 154-164, wherein the concentration of the surfactant or PEG is about 0.01%-3% w/v.
166. The composition of any one of Embodiments 154-164, wherein the concentration of the surfactant or PEG is about 0.05%-2% w/v.
167. The composition of any one of Embodiments 154-164, wherein the concentration of the surfactant or PEG is about 0.01%-3% v/v.
168. The composition of any one of Embodiments 154-164, wherein the concentration of the surfactant or PEG is about 0.05%-2% v/v.
169. The composition of any one of Embodiments 154-168, wherein the concentration of the buffer agent or buffer is about 0.1-10 mM.
170. The composition of any one of Embodiments 154-168, wherein the concentration of the buffer agent or buffer is about 0.2-5 mM.
171. The composition of any one of Embodiments 154-170, wherein the buffer agent is sodium phosphate monobasic.
172. The composition of any one of Embodiments 154-170, wherein the buffer agent is sodium phosphate monobasic.
173. The composition of any one of Embodiments 154-170, wherein the buffer is a phosphate buffer.
174. The composition of any one of Embodiments 154-170, wherein the buffer is a Tris buffer.
175. The composition of any one of Embodiments 154-174, wherein the pharmaceutically acceptable diluent is 5% Dextrose Injection, USP and the concentration of dextrose is about 0.02-0.05 mg/mL.
176. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
177. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

- a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
178. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
179. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
180. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
181. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

- PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
182. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
183. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water.
184. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
185. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

- PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
186. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
187. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
188. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
189. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
190. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

191. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

192. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

193. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

194. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

- arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
195. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water.
196. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
197. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
198. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

199. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
200. The composition of any one of Embodiments 176-199, wherein:
the concentration of I-66 is about 0.2-5 mg/mL;
the concentration of arginine is about 0.2-5 mg/mL;
the concentration of PEG-400 is about 0.06-1.7% w/v; and
the concentration of dextrose is about 0.02-0.05 mg/mL.
201. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
202. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
203. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.

204. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
205. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
206. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
207. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;

- optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
208. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water.
209. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
210. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
211. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
212. A composition, comprising:
I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

- a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.
213. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
214. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
215. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water.
216. A composition, consisting of:
I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

217. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

218. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

219. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water.

220. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.2-5 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water.

221. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

222. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

223. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

224. A composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL.

225. The composition of any one of Embodiments 176-199, wherein:

the concentration of I-66 is about 0.2-5 mg/mL;

the concentration of arginine is about 0.2-5 mg/mL;

the concentration of PEG-400 is about 0.06-1.7% v/v; and

the concentration of dextrose is about 0.02-0.05 mg/mL.

226. The composition of any one of Embodiments 176-225, wherein the buffer is a phosphate buffer.

227. The composition of any one of Embodiments 176-225, wherein the buffer is a Tris buffer.

228. The composition of any one of Embodiments 176-227, wherein the concentration of the buffer is about 0.01-10 mM.
229. The composition of any one of Embodiments 176-227, wherein the concentration of the buffer is about 0.02-5 mM.
230. The composition of any of the Embodiments 200-229, wherein the pH of the composition is about 7.0-9.0.
231. The composition of any of the Embodiments 200-229, wherein the pH of the composition is about 8.0.
232. The composition of any one of Embodiments 176-231, wherein I-66 is Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and the ¹³C NMR of I-66 in MeOD at 25°C, comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58, and I-66 has a purity of about or at least about 90%.
233. The composition of any one of Embodiments 176-232, wherein ¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23.
234. The composition of any one of Embodiments 176-233, wherein ¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise.

235. The composition of any one of Embodiments 176-234, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69.

236. The composition of any one of Embodiments 176-235, wherein ^1H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

237. The composition of any one of Embodiments 176-236, wherein identity of I-66 is confirmed by comparison with a reference standard.

238. The composition of Embodiment 237, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ^{13}C NMR in MeOD at 25°C, its ^{13}C NMR comprises the following peaks (chemical shift (δ , ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ^1H NMR in MeOD at 25°C, its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84,

1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

239. The composition of Embodiment 237 or 238, wherein the comparison is or comprising utilizing HPLC.

240. The composition of Embodiment 239, wherein the HPLC condition is selected from those in Example 1.

241. The composition of Embodiment 239, wherein the HPLC condition is as described in Table E-7.

242. The composition of Embodiment 239, wherein the HPLC condition is as described in Table E-10.

243. The composition of any one of Embodiments 237-242, wherein the comparison is or comprising utilizing NMR.

244. The composition of any one of Embodiments 237-243, wherein the comparison is or comprising utilizing ¹H NMR.

245. The composition of any one of Embodiments 237-244, wherein the comparison is or comprising utilizing ¹³C NMR.

246. The composition of any one of Embodiments 237-245, wherein the comparison is or comprising utilizing two- or multi-dimensional NMR.

247. The composition of any one of Embodiments 237-246, wherein the volume of the comparison is about 250 mL.
248. A composition, wherein the composition is identical with a composition of any one of the preceding Embodiments except that the concentration is v/v instead of w/v.
249. A composition, wherein the composition is identical with a composition of any one of Embodiments 1-247 except that the concentration is w/w instead of w/v.
250. The composition of any one of the preceding Embodiments, wherein I-66 has a purity of about 80% or more.
251. The composition of any one of the preceding Embodiments, wherein I-66 has a purity of about 85% or more.
252. The composition of any one of the preceding Embodiments, wherein I-66 has a purity of about 95% or more.
253. A method, comprising diluting a composition of I-66 with a pharmaceutically acceptable diluent.
254. The method of Embodiment 253, comprising:
calculating the volume of a composition of I-66 for administration of an intended dose;
removing from an infusion bag/container containing a pharmaceutically acceptable diluent a volume of the diluent equivalent to the calculated volume; and
addition of the calculated volume of the composition of I-66 to the infusion bag/container.
255. The method of any one of Embodiments 253-254, wherein the concentration of the I-66 in the composition is about 1-25 mg/mL.
256. The method of Embodiment 255, wherein the concentration of the I-66 in the composition is about 5-25 mg/mL.
257. The method of Embodiment 255, wherein the concentration of the I-66 in the composition is about 10 mg/mL.
258. The method of any one of Embodiments 253-257, wherein the composition of I-66 is a composition of any one of Embodiments 1-153.
259. The method of any one of Embodiments 253-257, wherein the composition of I-66 is a composition of any one of Embodiments 1-150.
260. The method of any one of Embodiments 253-259, wherein the pharmaceutically acceptable diluent is 5% Dextrose Injection, USP.
261. The method of any one of Embodiments 253-260, wherein the method prepares an I-66 for administration.
262. The method of any one of Embodiments 253-260, wherein the method prepares an I-66 for intravenous administration.
263. The method of any one of Embodiments 253-262, wherein the method prepares a composition of any one of Embodiments 154-252.

264. A method, comprising comparing I-66 in a preparation with a reference standard.

265. The method of Embodiment 264, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ¹H NMR in MeOD at 25°C, its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60,

1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

266. The method of Embodiment 264 or 265, wherein the comparison is or comprising utilizing HPLC.

267. The method of Embodiment 266, wherein the HPLC condition is selected from those in Example 1.

268. The method of Embodiment 266, wherein the HPLC condition is as described in Table E-7.

269. The method of Embodiment 266, wherein the HPLC condition is as described in Table E-10.

270. The method of any one of Embodiments 264-269, wherein the comparison is or comprising utilizing NMR.

271. The method of any one of Embodiments 264-270, wherein the comparison is or comprising utilizing ¹H NMR.

272. The method of any one of Embodiments 264-271, wherein the comparison is or comprising utilizing ¹³C NMR.

273. The method of any one of Embodiments 264-272, wherein the comparison is or comprising utilizing two- or multi-dimensional NMR.

274. The method of any one of Embodiments 264-273, wherein the method confirms identity of I-66 in the preparation.

275. The method of any one of Embodiments 264-274, wherein the method is utilized to control quality the I-66 preparation.

276. The method of any one of Embodiments 264-275, wherein the method is utilized to release the I-66 preparation.

277. The method of any one of Embodiments 264-276, wherein the method the I-66 preparation is a preparation of I-66 drug substance.

278. The method of any one of Embodiments 264-276, wherein the method the I-66 preparation is a preparation of I-66 drug product.

279. The method of any one of Embodiments 264-277, wherein I-66 in the preparation is in or comprises a pharmaceutically acceptable salt form.

280. The method of any one of Embodiments 264-279, wherein I-66 in the preparation is in or comprises a sodium salt form.

281. The method of any one of Embodiments 264-280, wherein I-66 in the reference is in or comprises a pharmaceutically acceptable salt form.

282. The method of any one of Embodiments 264-281, wherein I-66 in the reference is in or comprises a sodium salt form.

283. A method for modulating beta-catenin interaction with a partner in a system, comprising

administering or delivering to the system I-66.

284. The method of Embodiment 283, wherein the partner is TCF4.

285. A method for modulating a TCF-beta-catenin interaction in a system, comprising contacting beta-catenin with I-66.

286. A method for modulating a TCF-beta-catenin interaction in a system, comprising administering or delivering to the system I-66.

287. A method for inhibiting beta-catenin dependent cell proliferation, comprising administering or delivering to the system I-66.

288. A method for decreasing beta-catenin polypeptide level in a system, comprising administering or delivering to the system I-66.

289. A method for decreasing c-Myc polypeptide and/or transcript level in a system, comprising administering or delivering to the system I-66.

290. The method of any one of Embodiments 288-289, wherein c-Myc polypeptide level is reduced.

291. The method of any one of Embodiments 288-290, wherein c-Myc mRNA level is reduced.

292. The method of any one of Embodiments 288-291, wherein the reduction is observed 1, 2, 3, 4, 5, 6, 7, 8, 9, or 10 or more days post a dose.

293. A method for decreasing Axin2 polypeptide and/or transcript level in a system, comprising administering or delivering to the system I-66.

294. The method of any one of Embodiments 288-293, wherein Axin2 polypeptide level is reduced.

295. The method of any one of Embodiments 288-294, wherein Axin2 mRNA level is reduced.

296. A method for modulating WNT/beta-catenin pathway in a system, comprising administering or delivering to the system I-66, wherein expression of a nucleic acid is modulated.

297. A method, comprising administering or delivering to the system I-66, wherein level of a transcript of a nucleic acid and/or a product thereof is modulated.

298. A method, comprising administering or delivering to the system I-66, wherein expression of a nucleic acid is modulated.

299. The method of any one of Embodiments 296-298, wherein a nucleic acid is or comprises a gene.

300. The method of any one of Embodiments 283-299, wherein a system is an in vitro system.

301. The method of any one of Embodiments 283-299, wherein a system is an in vivo system.

302. The method of any one of Embodiments 283-299, wherein a system is or comprises a sample.

303. The method of any one of Embodiments 283-302, wherein a system is or comprises a cell, tissue or organ.

304. The method of any one of Embodiments 283-303, wherein a system is or comprises cancer cells.

305. The method of any one of Embodiments 283-305, wherein a system is or comprises a tumor.

306. The method of any one of Embodiments 283-305, wherein a system is a subject.

307. A method for treating cancer, comprising administering or delivering to a subject suffering therefrom

an effective amount of I-66.

- 308. A method, comprising administering or delivering to a subject suffering from cancer a dose of I-66.
- 309. The method of any one of Embodiments 307-308, wherein the cancer is a solid tumor.
- 310. The method of any one of Embodiments 307-308, wherein the cancer is colorectal cancer.
- 311. The method of any one of Embodiments 307-308, wherein the cancer is gastric cancer.
- 312. The method of any one of Embodiments 307-308, wherein the cancer is lung cancer.
- 313. The method of any one of Embodiments 307-308, wherein the cancer is non-small cell lung cancer.
- 314. The method of any one of Embodiments 307-308, wherein the cancer is metastatic non-small cell lung cancer.
- 315. The method of any one of Embodiments 307-308, wherein the cancer is non-small cell lung cancer stage IIIB.
- 316. The method of any one of Embodiments 307-308, wherein the cancer is non-small cell carcinoma.
- 317. The method of any one of Embodiments 307-308, wherein the cancer is non-small cell carcinoma of lung, TNM Stage 4.
- 318. The method of any one of Embodiments 307-308, wherein the cancer is gastroesophageal-junction cancer.
- 319. The method of any one of Embodiments 307-318, wherein the tumor is non-MSI-H.
- 320. The method of any one of Embodiments 307-318, wherein the tumor is non-dMMR.
- 321. The method of any one of Embodiments 307-320, wherein the cancer is a locally advanced cancer.
- 322. The method of any one of Embodiments 307-321, wherein the cancer is a metastatic cancer.
- 323. The method of any one of Embodiments 307-322, wherein the cancer is microsatellite stable.
- 324. The method of Embodiment 307, wherein the cancer is advanced or metastatic MSS colorectal cancer.
- 325. The method of Embodiment 307, wherein the cancer is advanced or metastatic non-small cell lung cancer.
- 326. The method of Embodiment 307, wherein the cancer is advanced or metastatic gastric cancer.
- 327. The method of Embodiment 307, wherein the cancer is advanced or metastatic gastroesophageal junction cancer.
- 328. The method of any one of Embodiments 307-327, wherein the cancer comprises a Wnt pathway activating mutation (WPAM).
- 329. The method of Embodiment 307, wherein the cancer is WPAM+ solid tumor.
- 330. The method of any one of Embodiments 307-329, wherein the cancer comprises an APC mutation.
- 331. The method of any one of Embodiments 307-330, wherein the cancer comprises a beta-catenin mutation.
- 332. The method of Embodiment 307, wherein the cancer is advanced or metastatic non-small cell lung cancer with documented WPAM in APC.

333. The method of Embodiment 307, wherein the cancer is advanced or metastatic non-small cell lung cancer with documented WPAM in beta-catenin.
334. The method of Embodiment 307, wherein the cancer is advanced or metastatic gastric cancer with documented WPAM in APC.
335. The method of Embodiment 307, wherein the cancer is advanced or metastatic gastric cancer with documented WPAM in beta-catenin.
336. The method of Embodiment 307, wherein the cancer is advanced or metastatic gastroesophageal junction cancer with documented WPAM in APC.
337. The method of Embodiment 307, wherein the cancer is advanced or metastatic gastroesophageal junction cancer with documented WPAM in beta-catenin.
338. The method of any one of Embodiments 307-337, comprising a mutation selected from *APC* loss of function mutations, *CTNNB1* gain of function mutations, *RNF43* LOF mutations, and *RSPO2* and *RSPO3* fusions.
339. The method of any one of Embodiments 307-338, comprising a mutation selected from mutations in *AMER1*, *AXIN1*, *AXIN2*, *BCL9*, *CSNK1A1*, *GSK3B*, *LRP5*, *LRP6*, *LGR5*, *TCF7L2*, and *WIF1*.
340. The method of any one of Embodiments 307-339, wherein cMyc expression in a tumor sample is reduced.
341. The method of any one of Embodiments 307-340, wherein the cancer is refractory.
342. The method of any one of Embodiments 283-341, comprising administering I-66 weekly.
343. The method of any one of Embodiments 283-342, comprising administering I-66 weekly for at least about three doses.
344. The method of any one of Embodiments 283-342, comprising administering I-66 weekly for at least about four doses.
345. The method of any one of Embodiments 283-344, comprising administering I-66 in a composition having a volume of about 250 mL.
346. The method of any one of Embodiments 283-345, comprising administering I-66 in a composition of any one of Embodiments 1-252.
347. The method of any one of Embodiments 283-345, comprising administering I-66 in a composition of any one of Embodiments 154-252.
348. The method of any one of Embodiments 283-345, comprising administering I-66 in a composition of any one of Embodiments 176-252.
349. The method of any one of Embodiments 283-348, comprising administering or delivering I-66 intravenously.
350. The method of any one of Embodiments 283-349, wherein a dose of about 18 mg/m² I-66 is administered.
351. The method of any one of Embodiments 283-350, wherein a dose of about 36 mg/m² I-66 is

administered.

352. The method of any one of Embodiments 283-351, wherein a dose of about 72 mg/m² I-66 is administered.

353. The method of any one of Embodiments 283-352, wherein a dose of about 144 mg/m² I-66 is administered.

354. The method of any one of Embodiments 283-353, wherein a dose of about 240 mg/m² I-66 is administered.

355. The method of any one of Embodiments 283-354, wherein a dose of about 360 mg/m² I-66 is administered.

356. The method of any one of Embodiments 283-355, wherein a dose of about 480 mg/m² I-66 is administered.

357. The method of any one of Embodiments 283-356, wherein a dose of about 600 mg/m² I-66 is administered.

358. The method of any one of Embodiments 283-357, comprising administering or delivering I-66 at assigned doses in continuous cycles.

359. The method of Embodiment 358, wherein the assigned dose of each cycle is about the same.

360. The method of any one of Embodiments 358-359, wherein the assigned dose of a cycle is different from another cycle.

361. The method of any one of Embodiments 358-360, wherein a cycle is about 21 days.

362. The method of any one of Embodiments 358-361, wherein a cycle is about 28 days.

363. The method of any one of Embodiments 358-360, wherein each cycle is independently about 21 days.

364. The method of any one of Embodiments 358-360, wherein each cycle is independently about 28 days.

365. The method of any one of Embodiments 358-364, wherein an assigned dose of a cycle is about 18 mg/m² I-66.

366. The method of any one of Embodiments 358-365, wherein an assigned dose of a cycle is about 36 mg/m² I-66.

367. The method of any one of Embodiments 358-366, wherein an assigned dose of a cycle is about 72 mg/m² I-66.

368. The method of any one of Embodiments 358-367, wherein an assigned dose of a cycle is about 144 mg/m² I-66.

369. The method of any one of Embodiments 358-368, wherein an assigned dose of a cycle is about 240 mg/m² I-66.

370. The method of any one of Embodiments 358-369, wherein an assigned dose of a cycle is about 360 mg/m² I-66.

371. The method of any one of Embodiments 358-370, wherein an assigned dose of a cycle is about 480 mg/m² I-66.
372. The method of any one of Embodiments 358-371, wherein an assigned dose of a cycle is about 600 mg/m² I-66.
373. The method of any one of the preceding Embodiments, comprising administering or deliver to a subject a second therapeutic agent.
374. The method of any one of the preceding Embodiments, comprising administering or deliver to a subject a second therapy.
375. The method of Embodiment 373 or 374, wherein a second therapeutic agent or therapy is administered prior to I-66.
376. The method of Embodiment 373 or 374, wherein a second therapeutic agent or therapy is administered concurrently with I-66.
377. The method of Embodiment 373 or 374, wherein a second therapeutic agent or therapy is administered subsequently to I-66.
378. The method of any one of the preceding Embodiments, wherein a subject is exposed to a second therapeutic agent or therapy and I-66.
379. The method of any one of the preceding Embodiments, wherein a subject is exposed to a therapeutic effect of a second therapeutic agent or therapy and a therapeutic effect of I-66.
380. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises a chemotherapy agent.
381. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises a hormone therapy agent.
382. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises an immunotherapy agent.
383. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises a checkpoint inhibitor.
384. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises an antibody.
385. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises a CTLA-4, PD-1 or PD-L1 inhibitor.
386. The method of any one of the preceding Embodiments, wherein a second therapeutic agent is or comprises a cell.
387. The method of any one of the preceding Embodiments, wherein the second therapeutic agent reduces one or more side effects of I-66.
388. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises surgery.

389. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises chemotherapy.

390. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises radiotherapy.

391. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises hormone therapy.

392. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises stem cell or bone marrow transplant.

393. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises immunotherapy.

394. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises T-cell therapy.

395. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises CAR T-cell therapy.

396. The method of any one of the preceding Embodiments, wherein a second therapy is or comprises administering to the subject a population of immune cells.

397. The method of any one of the preceding Embodiments, wherein the combination therapy provides higher efficacy than I-66 is administered or delivered alone.

398. The method of any one of the preceding Embodiments, wherein the combination therapy provides higher efficacy than when a second therapeutic agent or therapy is administered or delivered alone.

EXEMPLIFICATION

[0225] Those skilled in the art appreciate that various technologies are available for manufacturing and assessing provided technologies in accordance with the present disclosure. Described below are certain examples.

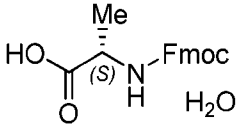
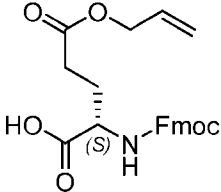
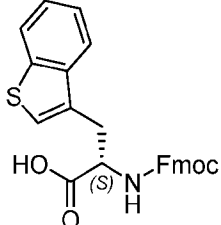
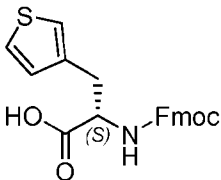
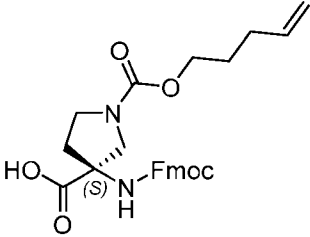
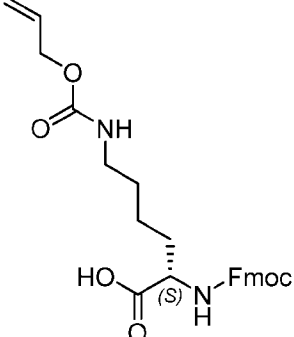
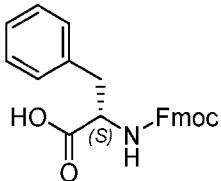
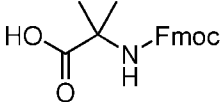
[0226] Example 1. Manufacturing of I-66.

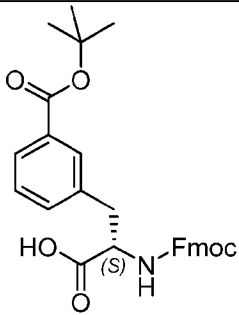
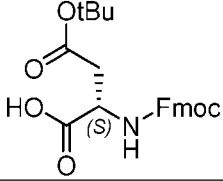
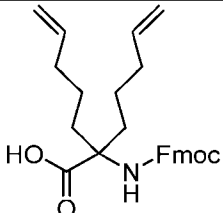
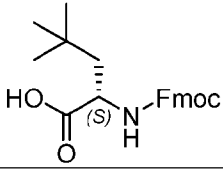
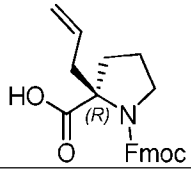
[0227] Those skilled in the art appreciate that various synthetic technologies may be utilized to prepare I-66 including drug substance and drug products in accordance with the present disclosure. Certain useful technologies are presented below as examples. Various technologies described in, e.g., WO2022/261257, are also useful for preparing I-66.

[0228] In some embodiments, the present disclosure provides technologies for manufacturing I-66, e.g., for use as a drug substance.

[0229] Certain useful starting materials are described below as examples.

Table E-1. Useful starting materials for I-66 preparation.

Name	Amino Acid Shorthand	Position in I-66	Structure
Fmoc-Ala-OH (hydrate)	Ala	15, 8	
Fmoc-Glu(OAllyl)-OH	Glu	14	
Fmoc-BztA-OH	BztA	13	
Fmoc-3Thi-OH	3Thi	12	
Fmoc-PyrS2-OH	PyrS2	11	
Fmoc-Lys(Alloc)-OH	Lys	10	
Fmoc-Phe-OH	Phe	9	
Fmoc-Aib-OH	Aib	7	

Name	Amino Acid Shorthand	Position in I-66	Structure
Fmoc-3COOHF(tBu)-OH	3COOHF	6	
Fmoc-Asp(OtBu)-OH	Asp	5, 2	
Fmoc-B5-OH	B5	4	
Fmoc-Npg-OH	Npg	3	
Fmoc-PL3-OH	PL3	1	

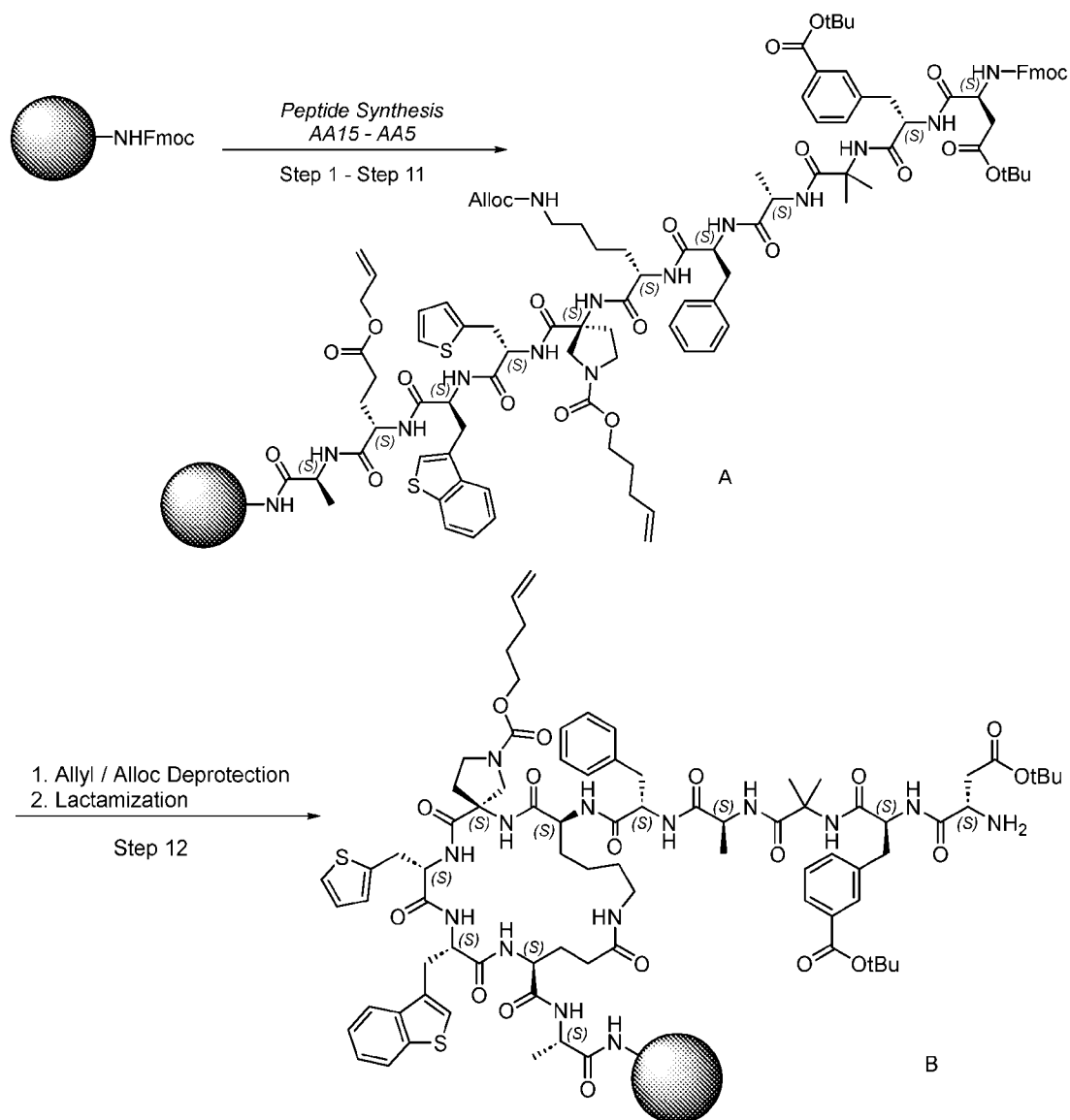
[0230] In some embodiments, I-66 is manufactured as described below as an example.

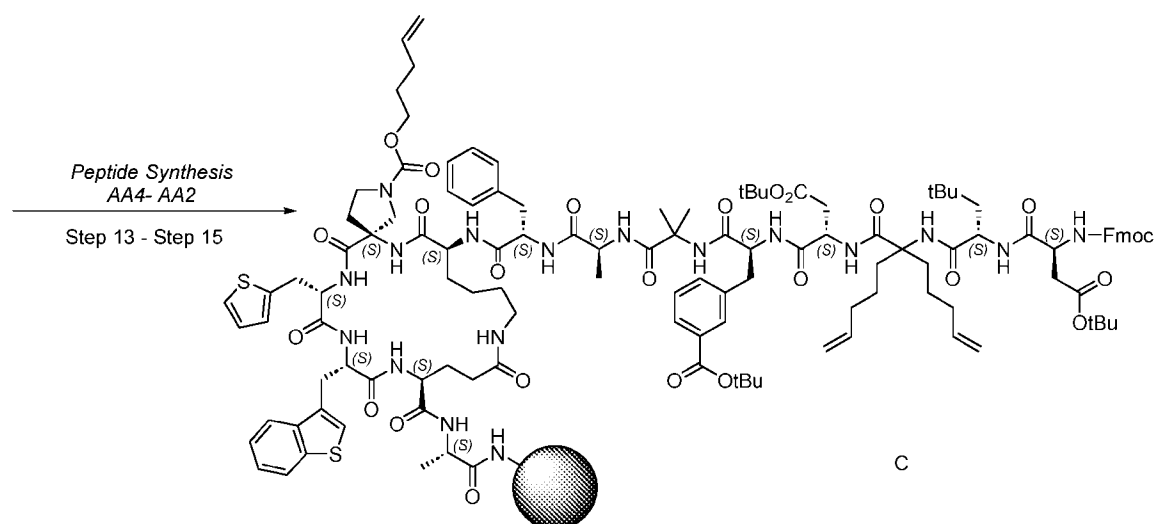
[0231] In some embodiments, I-66 drug substance manufacturing employs three stages. In the first stage, I-66 is synthesized using a solid-phase peptide synthesis (SPPS) approach with standard Fmoc protecting group strategy where all synthetic operations are performed on solid support, including installation of the olefin and lactam staples. Proceeding from the C-terminal to the N-terminal, synthesis is initiated by attaching the C-terminal amino acid residue onto the insoluble support (resin). After the initial attachment, subsequent Fmoc-protected amino acid derivatives are attached sequentially to lengthen the protected peptide anchored to resin. The second stage encompasses cleavage from resin. In the third stage, the crude peptide is purified by preparative reverse-phase chromatography and the product-bearing fractions are lyophilized to afford I-66 drug substance.

[0232] An overview of a synthetic process used for both the non-GMP batch for GLP toxicology studies as well as the GMP clinical batch, including the amide-forming steps and key stapling steps, is illustrated in

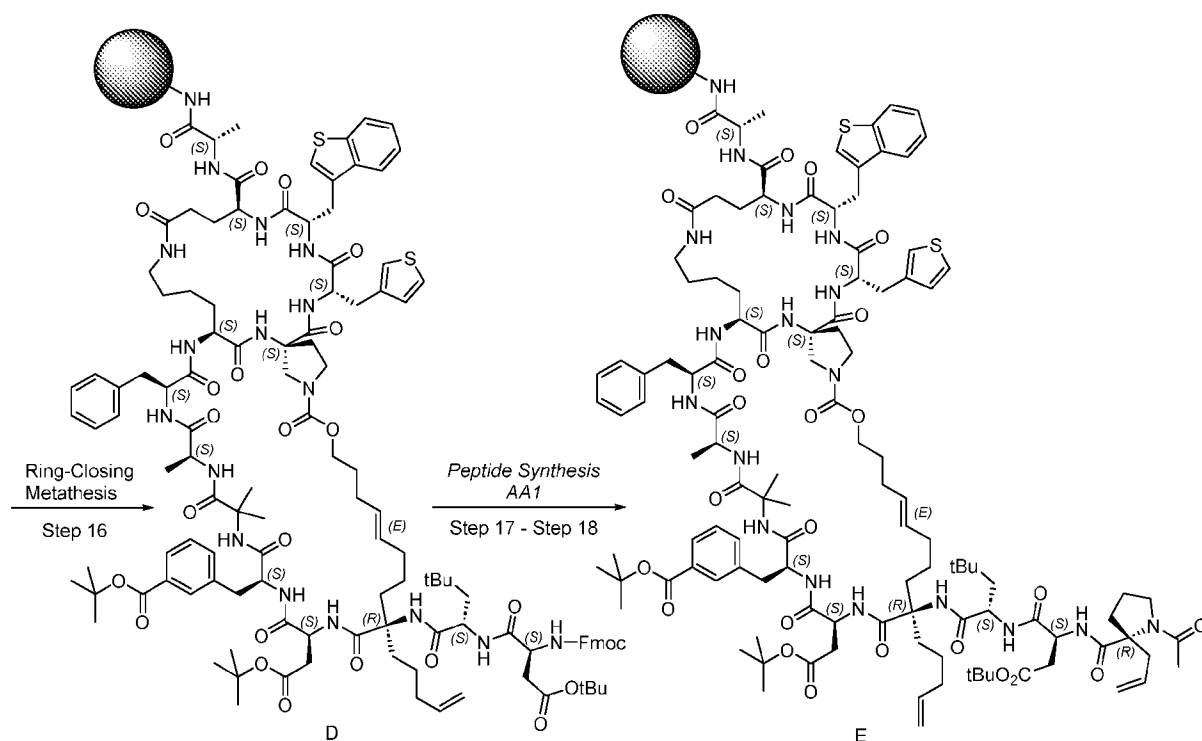
Scheme 1 and Scheme 2 below. Certain useful starting materials are described in Table E-1.

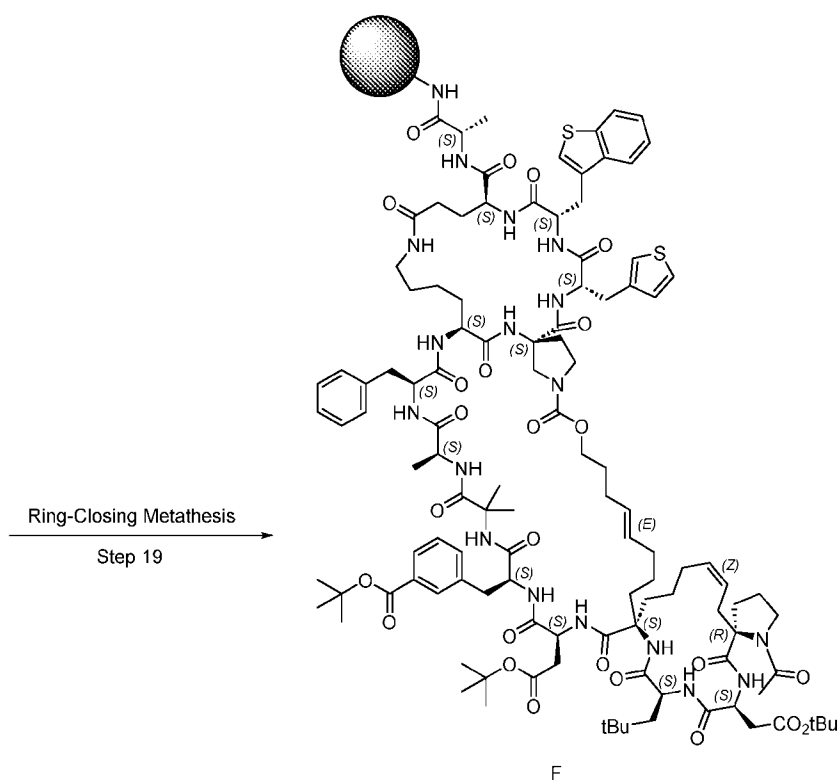
[0233] Scheme 1. SPPS Synthesis (Stage 1) of I-66 to AA2 Including First Staple Installation.





[0234] Scheme 2. Final Steps of I-66 SPPS Synthesis (Stage 1).

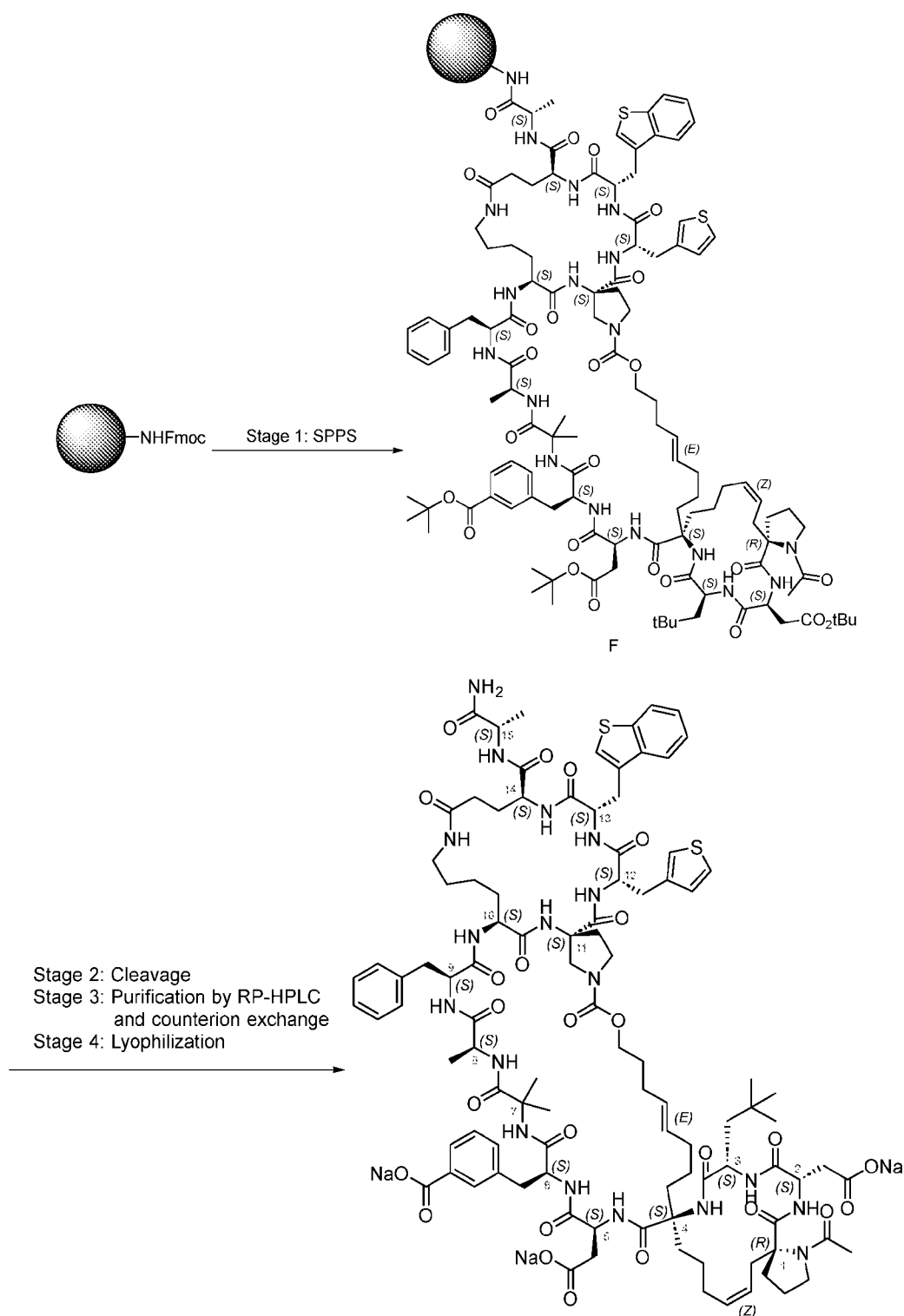




[0235] Typically, each peptide synthesis cycle step involves a deblocking, coupling, and capping step. The lactam stapling is performed after the first ten amino acids have been assembled. This is done by deprotection of the alloc and allyl groups on Lys¹⁰ and Glu¹⁴, respectively, using palladium-mediated deprotection conditions followed by amide bond formation using standard coupling agents (e.g., PyAOP, EDC, CDMT, etc.). Following additional elongation of the peptide chain to AA2, a first ring-closing metathesis to generate the (i,i+7) staple is performed (e.g., in various preparations, the *E*-olefin isomer). Then the last amino acid is installed, capped, and a second ring-closing metathesis is performed to generate the (i,i+3) staple (e.g., in various preparations, the *Z*-olefin isomer). Typically, one isomer is depicted, e.g., in schemes.

[0236] Following completion of the Stage 1 (SPPS) processing, I-66 is cleaved from the solid-support using TFA/water, neutralized and solid phase extracted using Sepabeads (Stage 2). The extracted peptide is recovered by filtration for the purification process. I-66 is purified by multi-step reverse-phase chromatography (Stage 3). The purified drug substance may be salt exchanged to provide the desired counterion before the purified solution is dried by lyophilization (Stage 4) to afford the substance before packaging in amber glass containers with Teflon-lined polypropylene screw caps and stored at $-20^{\circ}\text{C} \pm 5^{\circ}\text{C}$. A complete overview of an example manufacturing process is illustrated in Scheme 3 below.

[0237] Scheme 3. An Overview of All Manufacturing Stages.



[0238] Solid Phase Peptide Synthesis (SPPS) of the I-66 drug substance can be performed according to procedures and protocols well-developed and used routinely for peptide synthesis. SPPS typically comprises the sequential assembly of protected amino acid derivatives to synthesize a peptide anchored onto solid support (resin). In some embodiments, addition of an amino acid to grow the peptide generally involves the following steps: removal of protective N-terminal Fmoc group from the previous amino acid; wash; coupling

of the next activated amino acid; wash; acetylation (when necessary); and wash. Typically proceeding from the C-terminal to the N-terminal, these steps are repeated for each amino acid until the desired linear peptide is assembled onto resin.

[0239] In various preparations, at the start of the synthesis of I-66 drug substance, Rink-amide MBHA resin is prepared by a deblocking reaction using piperidine/dimethylformamide (DMF), followed by the loading of the C-terminal amino acid (Fmoc-Ala-OH·H₂O) onto resin in the presence of DMF/Oxymapure/diisopropylcarbodiimide (DIC). Unreacted sites on the resin are capped, e.g., by an acetylation reaction using DMF/diisopropylethylamine (DIPEA)/acetic anhydride. A substitution test is performed after the loading reaction to assure acceptable loading of the Fmoc-Ala-OH·H₂O onto resin. Each subsequent protected amino acid is added to grow the peptide by performing two reactions: [1] a deblocking reaction that removes the Fmoc group from the N-terminal amino acid derivative; and [2] a coupling reaction that attaches the next amino acid derivative. A capping reaction, e.g., an acetylation reaction is performed when necessary. These reactions are repeated iteratively to grow the peptide. The deblocking reaction is performed in the presence of piperidine/DMF. The coupling reaction is performed in the presence of Oxymapure/DIC/DMF. The acetylation reaction is performed in the presence of DMF/DIPEA/acetic anhydride. In-process checks (e.g., Ninhydrin test or chloranil test, and/or LC analysis) are performed after each reaction to assure the completeness of each reaction. Wash steps are applied at the end of each reaction to remove residual reaction reagents and solvents, and to prepare for the next reaction.

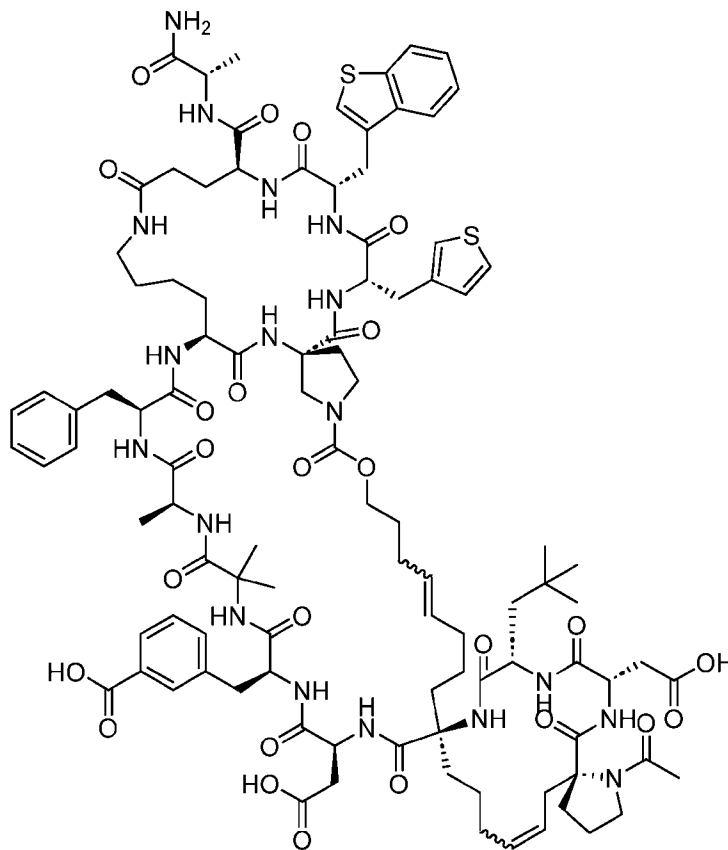
[0240] After the attachment of Fmoc-Asp⁵(OtBu)-OH, an allylic deprotection reaction followed by a lactamization reaction are performed to bridge Lys¹⁰ and Glu¹⁴. The allylic deprotection reaction is performed in the presence of tetrakis(triphenylphosphine palladium (Pd(PPh₃)₄)/triphenylphosphine (PPh₃)/dimethylbarbituric acid (DMBA)/dichloromethane (DCM). The lactamization reaction is performed in the presence of (7-Azabenzotriazol-1-yloxy)tripyrrolidinophosphonium hexafluorophosphate (PyAOP)/DMF/DIPEA. In-process checks are performed after each reaction to assure the completeness of each reaction. After the lactamization reaction, synthesis of the peptide resumes.

[0241] After the attachment of Fmoc-Asp²(OtBu)-OH, a ring closing metathesis (RCM) reaction is performed to connect B5⁴ to PyrS2¹¹, in the presence of Hoveyda-Grubbs catalyst. In-process checks were performed to assure the completeness of each reaction. Synthesis of the peptide resumes with the attachment of Fmoc-PL3¹-OH and its acetylation at the N-terminal. A second RCM reaction is performed to connect B5⁴ to PL3¹. The peptide-resin is dried under reduced pressure and elevated temperature.

[0242] Cleavage of peptide from resin and Solid Phase Extraction (SPE)

[0243] Crude I-66 drug substance is cleaved from resin and deprotected by removal of the side-chain protective groups by acidolysis, in the presence of trifluoroacetic acid (TFA)/H₂O. Spent resin is removed by filtration, the filtrate is neutralized, and subjected to solid phase extraction (SPE) using Sepabeads resin in the presence of ACN/NH₄OAc. In-process checks are performed to assure the completeness of the extraction. The resin is collected and washed. The crude peptide is released from resin in the presence of ACN/H₂O and

separated from spent resin by filtration for the purification steps. Various stereoisomer may be formed; I-66 is the isomer that show various characterization data (e.g., NMR, HPLC, etc.) as described herein and is purified.



[0244] Purification by reverse phase chromatography

[0245] Crude I-66 drug substance is purified by a three-step reverse phase chromatography (RPC) process (e.g., RPC 1, RPC 2, and RPC 3). The crude peptide (in a solution) is diluted 1:4 with ACN/ NH_4HCO_3 , then filtered through 2.4 μm filter and applied to the prepared HPLC column. Effluent from the column is monitored for UV absorption and fractions are collected when the product begins to elute. The fractions are assessed by analytical HPLC and pooled in such a way that the purity complies with the requirements for in-process controls. The fractions meeting the purity requirements are pooled for the next purification step. Side fractions not meeting the purity requirements are either pooled to a repeat of the purification steps or discarded.

[0246] The first purification step (RPC 1) is by preparative HPLC using a column of reverse phase media that has been washed and pre-equilibrated with ACN/ NH_4HCO_3 . The adsorbed peptide is eluted from the column by applying a gradient of ACN/ NH_4HCO_3 . Fractions are collected when the product starts to elute from the column and analyzed for peptide purity by analytical HPLC. RPC 1 fractions meeting the specified in-process control (IPC 1) acceptance criteria for purity (e.g., $\geq 85\%$) are pooled for RPC 2 and kept at 2 – 8°C until RPC 2. RPC 1 side fractions not meeting the purity acceptance criteria are either pooled to repeat

RPC 1 or discarded.

[0247] The main pools from RPC 1 are diluted with an equal volume of NH_4HCO_3 and applied to the preparative HPLC column equilibrated with $\text{ACN}/\text{NH}_4\text{HCO}_3$. The adsorbed peptide is eluted from the column by applying a gradient of $\text{ACN}/\text{NaH}_2\text{PO}_4$, pH 8.0. Fractions are collected when the product starts to elute from the column and analyzed for peptide purity by analytical HPLC. RPC 2 fractions meeting the in-process control (IPC 2) acceptance criteria for purity (e.g., $\geq 94\%$ Pre-pool and $\geq 95\%$ Main pool) are pooled for RPC 3 and kept at $2 - 8^\circ\text{C}$ until RPC 3. RPC 2 side fractions not meeting the purity acceptance criteria are either pooled to repeat RPC 2, pooled to repeat RPC 1, or discarded. The pooled side fractions are kept at $2 - 8^\circ\text{C}$ until their respective next steps.

[0248] The main pools from RPC 2 are diluted with an equal volume of USP Purified water and applied to the preparative HPLC column equilibrated with $\text{ACN}/\text{NaH}_2\text{PO}_4$, pH 8.0. The adsorbed peptide is eluted from the column by applying a gradient of $\text{ACN}/\text{NH}_4\text{HCO}_3$. Fractions are collected when the product starts to elute from the column and analyzed for peptide purity by analytical HPLC. RPC 3 fractions meeting the in-process control (IPC 3) acceptance criteria for purity (e.g., $\geq 94.5\%$ Pre-pool and $\geq 95.0\%$ Main pool) are pooled for the sodium conversion step and kept at ambient temperature until the sodium conversion step. RPC 3 side fractions not meeting the purity acceptance criteria are either pooled to repeat RPC 2, pooled to repeat RPC 1, or discarded. The pooled side fractions are kept at $2 - 8^\circ\text{C}$ until their respective next steps.

[0249] Sodium conversion and finished product isolation

[0250] The main pools from RPC 3 are combined, diluted 1:1 with Sterile Water for Irrigation, USP to the target peptide concentration range, and added to an appropriate amount of filtered sodium hydroxide solution required to convert the product to its trisodium form. The sodium form of the product is filtered through a $0.2\text{ }\mu\text{m}$ filter for final lyophilization.

[0251] After final lyophilization is completed, the finished I-66 drug substance (in powder form) is packaged in amber glass containers, capped with Teflon®-lined polypropylene screw caps, and stored at $-20^\circ\text{C} \pm 5^\circ\text{C}$.

[0252] In some cases, lyophilized I-66 drug substance that did not meet a desired acceptance criteria as established in the desired drug substance specification can be subjected to re-purification. Re-purification is carried out, after reconstitution of the peptide in an appropriate solvent, by repeating the purification step(s), described above. After re-purification, the material obtained undergoes final lyophilization as described above.

[0253] In some embodiments, lyophilized I-66 drug substance not meeting a desired acceptance criteria for residual solvents, salts, or water content established in the peptide drug substance specification may be subjected to re-lyophilization. Re-lyophilization is carried out, after reconstitution of the peptide, by repeating the lyophilization step, as described above.

[0254] In some embodiments, the following method is utilized for assessing I-66 purity; in some embodiments, it is utilized for in-process control of preparative HPLC fractions.

[0255] Table E-2. A Useful HPLC Protocol.

Parameter	Description	
Column	Acquity CSH C18 130 Å, 1.7 µm, 2.1 mm x 150 mm	
Column Temperature	45 °C	
Flow Rate	0.25 mL/min	
Detector Wavelength	220 nm	
Buffer A	0.1% TFA in Water	
Buffer B	0.1% TFA in Acetonitrile	
Gradient	Time (min)	%B
	0	50
	40	90
	48	90
	48.1	50
	60.1	50

[0256] Other in-process controls being used during the manufacturing process include monitoring to assure completion of each of the synthetic coupling reactions during the assembly of the protected peptide-resin precursor using Ninhydrin and/or Chloranil tests and/or RP-HPLC analysis; verifying purity/impurity profile of the product by RP-HPLC following the cleavage of the crude product from the resin; and verifying identity of the product by LC-MS following the cleavage of the crude product from the resin.

[0257] In some embodiments, I-66 was isolated as the sodium salt and isolated by lyophilization as an amorphous solid.

[0258] Characterization

[0259] I-66 is characterized by various technologies. Certain characterizations are presented as examples.

[0260] Ultra-High Performance Liquid Chromatography-Mass Spectrometry (UHPLC-MS) can be used to verify the monoisotopic mass of the I-66 drug substance. A sample was analyzed on a Waters Xevo G2-XS QToF mass spectrometer to confirm the monoisotopic mass of a preparation. The theoretical monoisotopic mass is 2074.93 Da (as C₁₀₂H₁₃₄N₁₈O₂₅S₂). The charged molecular ion [M+2H]²⁺ was observed during a mass spectral analysis: m/z calculated 1038.4677; observed 1038.47. The results confirm the theoretical molecular mass of I-66.

[0261] Matrix-assisted Absorption and Desorption/Ionization-Time-of-flight tandem mass spectrometry (MALDI-TOF MS/MS) can be used to verify the amino acid sequence of I-66. A sample was analyzed on a Bruker ultrafleXtreme MALDI-TOF Mass Spectrometer to confirm the monoisotopic mass and the peptide sequence. The results confirm the amino acid sequence of I-66.

[0262] Chiral amino acid analysis (e.g., using GC/MS) can be performed to determine chiral purity of various constituent amino acid residues, e.g., Ala, Npg, Asp, Phe, Lys, and BztA. In an analysis, the peptide is hydrolyzed in 6N deuterated solvents (DCI/D₂O), suitably derivatized, and analyzed using GC-MS to determine each amino acid enantiomer. Racemization during hydrolysis and derivatization (sample prep) is paralleled by deuterium exchange in the α-position (deuterium label). The proportion of epimerized-amino

acid present in the I-66 sample is represented by the relative amounts of the unlabeled form which is monitored by mass spectrometry. The results of the analysis confirm that the chiral and isomeric purity of all the standard L-amino acids present in I-66 drug substance is high.

[0263] Table E-3. Amino Acid Enantiomeric Purity from An Analysis.

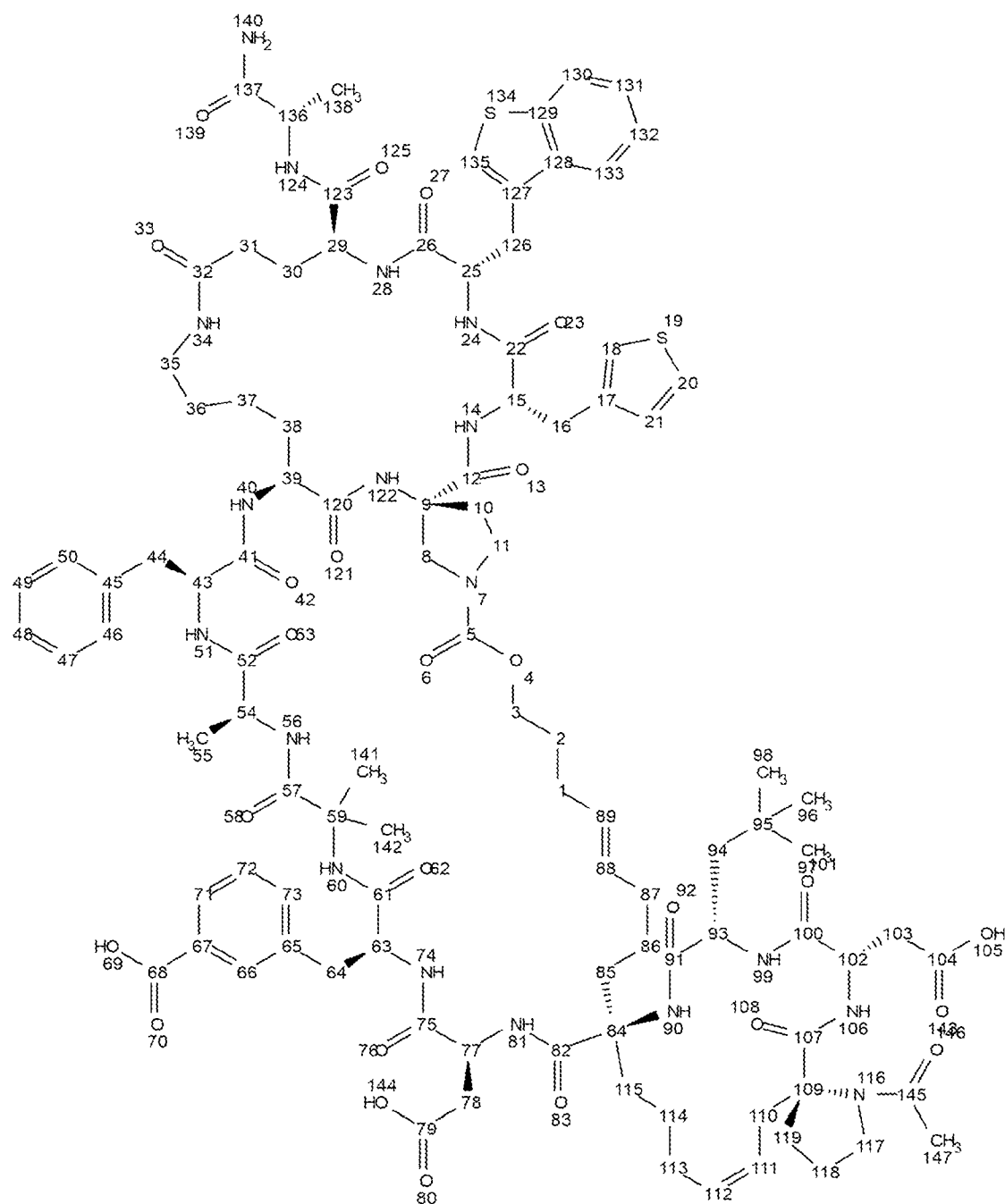
Amino acid	Enantiomeric purity (% L-enantiomer)
Alanine (Ala)	≥ 99.90
β -tBu-Alanine (Npg)	99.86
Aspartic Acid (Asp)	≥ 99.90
Phenylalanine (Phe)	≥ 99.90
Lysine (Lys)	≥ 99.90
Benzothienylalanine (BztA)	99.77

[0264] In some embodiments, the present disclosure provides I-66 preparations with chiral amino acid purities about as described above. In some embodiments, enantiomeric purity of Ala is $\geq$ about 99.9%, enantiomeric purity of Npg is $\geq$ about 99.8%, enantiomeric purity of Asp is $\geq$ about 99.9%, enantiomeric purity of Phe is $\geq$ about 99.9%, enantiomeric purity of Lys is $\geq$ about 99.9%, and/or enantiomeric purity of BztA is $\geq$ about 99.7%.

[0265] Preparations of I-66 may contain counterions. In some embodiments, sodium and ammonium content of batches manufactured was assessed by ion chromatography to confirm the identity and determine the quantity of the counter ion. For preparations prepared as sodium salt, identity of the sodium counter ion was confirmed which corresponds to approximately 3 equivalents per molecule and ammonium was not detected. For a preparation prepared as ammonium salt before lyophilization, ammonium content was below the theoretical level for the stoichiometric salt, indicating a possible disproportionation of the salt during lyophilization.

[0266] ^1H - and ^{13}C Nuclear Magnetic Resonance Spectroscopy (NMR) technologies are utilized to confirm I-66 identity and assess I-66 preparations. For example, for a preparation, sample was dissolved in methanol- d_4 . ^1H , ^{13}C , COSY, TOCSY, NOESY, HSQC and HMBC data were acquired on a 900MHz Bruker AVANCE III system with a cryoprobe. Sample concentration was 10mg/750 μl . Observed ^1H and ^{13}C signals are presented in the third and fourth columns in Table E-4. Without the intention to limited by them, possible assignments were also included in Table E-4 and the first and second columns.

[0267] Table E-4. ^1H and ^{13}C Signals and Possible Assignments (MeOD at 25°C)



Atom number	Atom type	H chemical shift (δ , ppm)	C chemical shift (δ , ppm)
1	CH ₂	2.00, 2.10	29.99
2	CH ₂	1.60, 1.65	30.63
3	CH ₂	3.73, 4.15	67.18
4	O	-	-
5	C=O	-	156.51
6	O	-	-
7	N	-	-
8	CH ₂	3.15, 4.22	53.94
9	C	-	65.89
10	CH ₂	2.29, 2.41	31.81

Atom number	Atom type	H chemical shift (δ , ppm)	C chemical shift (δ , ppm)
11	CH ₂	3.53, 3.25	45.37
12	C=O	-	173.13
13	O	-	-
14	NH	8.32	-
15	CH	3.96	59.94
16	CH ₂	3.06, 2.95	32.60
17	C	-	137.36
18	CH	6.34	123.42
19	S	-	-
20	CH	6.84	125.19
21	CH	6.36	129.15
22	C=O	-	175.55
23	O	-	-
24	NH	7.73	-
25	CH	4.38	56.93
26	C=O	-	175.8
27	O	-	-
28	NH	8.17	-
29	CH	4.31	55.69
30	CH ₂	1.93	27.47
31	CH ₂	2.38	33.84
32	C=O	-	174.47
33	O	-	-
34	NH	-	-
35	CH ₂	2.92, 3.45	39.25
36	CH ₂	1.18	23.2
37	CH ₂	1.36, 1.55	35.8
38	CH ₂	1.84, 1.53	29.61
39	CH	3.82	57.70
40	NH	8.19	-
41	C=O	-	175.8
42	O	-	-
43	CH	4.21	60.70
44	CH ₂	3.27, 3.31	38.18
45	C	-	138.13
46	CH	7.21	130.39
47	CH	7.24	129.83
48	CH	7.07	128.08
49	CH	7.24	129.83
50	CH	7.21	130.39
51	NH	8.25	-
52	C=O	-	177.56
53	O	-	-
54	CH	4.03	54.54
55	CH ₃	1.55	17.00
56	NH	8.44	-
57	C=O	-	178.58
58	O	-	-
59	C	-	57.28
60	NH	8.89	-

Atom number	Atom type	H chemical shift (δ , ppm)	C chemical shift (δ , ppm)
61	C=O	-	174.39
62	O	-	-
63	CH	4.25	60.94
64	CH ₂	3.27, 3.44	38.10
65	C	-	138.06
66	CH	7.97	131.96
67	C	-	132.12
68	C	-	169.51
69	OH	-	-
70	O	-	-
71	CH	7.94	129.37
72	CH	7.42	129.54
73	CH	7.50	135.08
74	NH	8.10	-
75	C=O	-	175.7
76	O	-	-
77	CH	4.40	55.56
78	CH ₂	2.74, 2.96	36.18
79	HO-C=O	-	172.48
80	O	-	-
81	NH	8.42	-
82	C=O	-	177.0
83	O	-	-
84	C	-	62.78
85	CH ₂	1.92, 2.07	30.00
86	CH ₂	1.37	23.32
87	CH ₂	2.38	33.8
88	CH	5.29	131.26
89	CH	5.31	131.47
90	NH	7.45	-
91	C=O	-	175.8
92	O	-	-
93	CH	4.27	56.48
94	CH ₂	1.80, 1.88	45.96
95	C	-	31.36
96, 97, 98	CH ₃	1.07	30.18
99	NH	7.39	-
100	C=O	-	175.44
101	O	-	-
102	CH	4.31	55.33
103	CH ₂	2.71, 2.81	35.69
104	HO-C=O	-	174.10
105	OH	-	-
106	NH	8.15	-
107	C=O	-	175.6
108	O	-	-
109	C	-	70.51
110	CH ₂	3.20, 2.05	33.09
111	CH	5.56	129.2
112	CH	5.36	126.96

Atom number	Atom type	H chemical shift (δ , ppm)	C chemical shift (δ , ppm)
113	CH ₂	1.97, 2.17	27.46
114	CH ₂	0.84, 1.42	23.29
115	CH ₂	1.57, 2.69	35.96
116	N	-	-
117	CH ₂	3.76, 3.92	49.50
118	CH ₂	2.20	24.46
119	CH ₂	2.42, 2.22	36.46
120	C=O	-	176.97
121	O	-	-
122	NH	8.86	-
123	C=O	-	175.55
124	NH	7.69	-
125	O	-	-
126	CH ₂	3.08, 3.40	31.76
127	C	-	132.57
128	C	-	139.58
129	C	-	141.98
130	CH	7.89	122.71
131	CH	7.38	125.37
132	CH	7.41	125.20
133	CH	7.89	123.87
134	S	-	-
135	CH	7.00	126.99
136	CH	4.30	51.20
137	C=O	-	178.08
138	CH ₃	1.42	17.76
139	O	-	-
140	NH ₂	-	-
141	CH ₃	1.59	28.98
142	CH ₃	1.33	23.20
143	O	-	-
144	OH	-	-
145	C=O	-	172.38
146	O	-	-
147	CH ₃	2.23	23.87

[0268] There is a strong and unambiguous NOE peak observed between the olefin protons 111 and 112. This peak is stronger than the NOE peak between neighboring aromatic protons 20 and 21, or 48 and 47/49. Without the intention to be limited by theory, since the distance between neighboring aromatic protons is about 2.5Å, the distance between 111 and 112 is likely shorter than 2.5Å. The two protons are drawn as *cis* configuration in various chemical structures of the present disclosure to represent these observations. Both proton and carbon peaks for 88 and 89 have similar chemical shift values on a 900MHz instrument. Traditional NOE experiments have not provided as much information about stereochemistry between the two protons. A combination of homonuclear decoupling experiment and simulation showed that the coupling constant between protons 88 and 89 is about 14Hz. The two protons are drawn as *trans* configuration in various chemical structures of the present disclosure to represent these observations. While stereoisomers

exist, those skilled in the art readily appreciate that identity of I-66 is established and confirmed by various data herein, e.g., NMR, HPLC, etc.

[0269] X-ray crystallography was used to characterize the structure of I-66. The obtained data confirm the helical nature of I-66 as it binds to β -catenin. X-ray also confirmed the primary sequence of I-66.

[0270] Secondary structure of I-66 was analyzed using Microfluidic Modulation Spectroscopy (MMS). I-66 at 20 mg/mL was analyzed in its formulation (10 mg/mL arginine; 33.6 mg/mL PEG-400; 1.2 mg/mL sodium phosphate monobasic; pH 8) on an AQS3@pro MMS production system (RedShiftBio, Boxborough, MA). The absolute absorbance spectrum and its second derivative plot of the peptide of a batch are shown in Figure 14, (A) and (B), respectively. The major peak at 1662 cm^{-1} indicates that the secondary structure of the protein is dominated by a helical structure, with a mixed of α - and 3_{10} -helix characters. This agrees with the crystal structure of I-66. Quantitative analysis of the Higher Order Structure (HOS) fractional contributions (Figure 14, (C)) was performed by Gaussian curve deconvolution of the baseline-corrected second derivative plot, based on the secondary structural element designations shown below:

Wavenumber (cm^{-1})	Secondary Structural Element
1612	Extended strand
1632	β -sheet
1638	β -sheet
1650	Unordered
1656	α -helix
1663	3_{10} -helix
1672	Turn
1680	Turn
1688	Turn

[0271] As shown in Figure 14, (C), roughly 56% of the structure of the batch is 3_{10} -helix and 13% of the structure is α -helix, totaling 69% of helical content. The total percentage of helical structure also agrees with the crystal structure, showing 12 out of the 18 (67%) amide carbonyl groups are forming H-bonds responsible for the helical structure. The relatively higher 3_{10} -helix content compared to α -helix can be explained by the crystal structure, which shows that some of the amide carbonyls have the potential to H-bonds (based on the calculated distance) with both the i+3 and i+4 amines, which can result in a mix of both 3_{10} - and α -helix characters. In solution, where the MMS measurements were taken, these H-bonding interactions can be dynamic and possibly shift to one character more than the other depending on the level of hydration. Examples of mixed α -/ 3_{10} -helix characters and the extended helix strand in an I-66 crystal structure include H-Bond formation between Ac⁰ and Npg³/B5⁴, H-Bond formation between Npg³ and 3COOHF⁶/Aib⁷, and residues 3Thi¹², BztA¹³, and Glu¹⁴ in the extended strand.

[0272] In certain preparations, I-66 was isolated by lyophilization and no crystalline or polymorphic forms were observed. For example, a sample of a preparation (sodium salt) was assessed by XRPD using a Panalytical Empyrean diffractometer equipped with a Cu X-ray tube and a PIXcel 1D-Medipix3 detector system at ambient temperature in transmission mode and held between low density polyethylene films. The

following parameters were used: range 3-40°2 θ , step size 0.01313°, counting time 48sec, ~10min run time, counting time 23sec, ~5min run time. Samples were spun at 60 rpm during data collection. XRPD patterns were sorted and manipulated using HighScore Plus v4.9 software. The sample was found to be amorphous with no clear indication of crystallinity.

[0273] Thermal analysis can be utilized to assess I-66 preparations. For example, Hyper-DSC was carried out to determine the glass transition temperature of an I-66 preparation. The thermogram did not detect the glass transition temperature of the amorphous material. Hyper DSC analyses were carried out on a Perkin Elmer DSC8500 Differential Scanning Calorimeter. Accurately weighed samples were placed in crimped aluminum pans, closed but not gas tight. Each sample was heated and cooled under nitrogen over two cycles at a rate of 300 °C/minute using a set temperature range of -50 to 160 °C. Indium metal was used as the calibration standard.

[0274] Thermogravimetric differential thermal analysis (TG-DTA) can be utilized to assess I-66 preparations. For a preparation, thermogravimetric analysis was carried out on a preparation and showed significant weight losses, likely due to loss of volatile components. A 9% weight loss from 60-170°C which equates to 11.7 mols of water was observed. Melting points were not detected in the DTA traces, in keeping with amorphous behavior. Thermogravimetric analyses were carried out on a Mettler Toledo TGA/DSC1 STARe. The calibration standards were indium and tin. Samples were placed in an aluminum sample pan, inserted into the TG furnace and accurately weighed. Under a stream of nitrogen at a rate of 10°C/minute, the heat flow signal was stabilized for one minute at 30°C, prior to heating to 300°C.

[0275] Dynamic Vapor Sorption (DVS) can be utilized to assess I-66 preparations. For example, the hygroscopicity and the sorption properties of an I-66 preparation were determined using Dynamic Vapor Sorption (DVS). The sample exhibited continuous uptake of moisture, generating a steady increase in weight from 40-90% RH. The isothermal plot showed the total weight gain observed between ambient (40% RH) and 80% RH was 13.3% w/w, which indicated that the sample is hygroscopic, based on the European Pharmacopoeia classification. The rate of desorption was comparable with that of sorption, with little hysteresis. Post DVS XRPD analysis was carried out and the resulting XRPD pattern matched that of the supplied material, indicating that there were no apparent physical changes. Dynamic Vapor Sorption (DVS) was performed using a SMS DVS Intrinsic Vapor Sorption Balance. Approximately 30mg of sample was placed into an aluminum balance pan, loaded into the vapor sorption balance and held at 25°C $\pm$ 0.1°C. The sample was subjected to a 2-cycle step profile in 10% increments from 40-90-0-90-0-40%RH. The equilibrium criterion was set as $\partial m / \partial t = 0.002\%/min$ with a minimum of 60 minutes and a maximum of 5 hours for each increment. The weight change during the sorption cycle was monitored, allowing for the hygroscopic nature of the sample to be determined. The data collection interval was in seconds.

[0276] The specific optical rotation for a preparation of I-66 was determined for I-66 according to USP <781S> as $[\alpha]_D^{25} = -73.4^\circ$ ($c = 0.1\%$ in 1% acetic acid / water).

[0277] Dissociation Constant and Isoelectric point. One pKa value for an I-66 preparation was

measured at 5.0. Precipitation prevented determination of further pKa values. The calculated theoretical isoelectric point for I-66 is 5.0.

[0278] Ultraviolet-Visible Spectroscopy (UV-Vis). The ultraviolet and visible (UV-Vis) spectrum of I-66 at a concentration of 0.05 mg/mL (24 mM) showed absorbance peaks at about 200 nM and about 230 nM. At varying concentrations in methanol, absorbance maxima / relevant absorbances and corresponding molar absorptivities are listed below.

[0279] Table E-5. UV-Vis Spectrum Results for I-66 in Methanol (190-800 nm).

Absorbance maxima wavelength (nm)	Conc. (mM)	Absorbance (AU)	Molar Absorptivity (L/mol•cm)
200	0.024	0.556	2.3×10^5
230	0.048	0.511	1.1×10^5
290	0.48	0.145	3.0×10^3
300	0.48	0.105	2.2×10^3

Path length: 1 mm

[0280] Solubility Profile: The solubility of I-66 was estimated in 18 solvent systems using the aliquot addition method. These included 4 aqueous/organic mixtures. I-66 had a solubility of >25mg/mL in 9 of the solvents and 3 of the aqueous mixtures at ambient temperature. Solubility data obtained are shown below as examples. Those experiments which did not show dissolution in ~40 volumes were temperature cycled for 2 cycles.

[0281] Table E-6. Solubility Estimates of I-66 Free Acid at 20°C.

Solvent	Acronym	Solubility Range (mg/mL)
acetone	-	<27
acetonitrile	ACN	<26
dichloromethane	DCM	<10
dimethyl sulfoxide	DMSO	>198
dioxane	-	<10
ethyl acetate	EtOAc	<24
hexafluoroisopropanol	HFIPA	>204
methanol	MeOH	>408
methyl ethyl ketone	MEK	<10
methyl tert-butyl ether	MTBE	<10
isopropanol	IPA	27-32
tetrahydrofuran	THF	26-30
water	-	<26
Water-IPA (1:1)	-	42-52
Water-ACN (1:1)	-	50-66
Water-THF (1:1)	-	102-204
0.1M NaOH pH = 13	-	63-95
Water-acetone (1:1)	-	<10

[0282] All batches of I-66 manufactured for assessment are of high purity, with a GMP purity specification of $\geq 95\%$. In some embodiments, purity of an I-66 preparation is about 93% or more. In some embodiments, purity of an I-66 preparation is about 94% or more. In some embodiments, purity of an I-66

preparation is about 95% or more. In some embodiments, an I-66 preparation comprises one or more I-66 isomers. In some embodiments, an I-66 preparation comprises one or more peaks with RRTs close to I-66 and with the same mass as I-66. For example, without the intention to be limited by theory, in some embodiments, close-eluting impurities to I-66 (e.g., RRT 1.02, 1.04; RRT of I-66 is 1.00) have been identified as olefin isomer impurities formed during the ring-closing metathesis steps of the (i,i+3) and (i,i+7) staples. In some embodiments, a preparation or composition comprises a fraction with a RRT of 1.02 and the same mass as I-66, and a fraction with a RRT of 1.04 and the same mass as DS-2. In some embodiments, each fraction is independently of a single component. In some embodiments, the RRT 1.02 fraction is about or no more than about 1.5%-2.5%, e.g., about or no more than about 1.5%, 1.6%, 1.7%, 1.8%, 1.9%, 2.0%, 2.1%, 2.2%, 2.3%, or 1.8%-2.3%, etc. In some embodiments, the RRT 1.04 fraction is about or no more than about 0.9%-3.0%, e.g., about or no more than about 1.0%, 1.1%, 1.2%, 1.3%, 1.4%, 1.5%, 1.6%, 1.7%, 1.8%, 1.9%, 2.0%, 2.1%, 2.2%, 2.3%, 2.4%, 2.5%, 2.6%, 2.7%, 2.8%, 2.9%, 3.9%, 0.9%-2.9%, 2.0%-3.0%, etc.

[0283] In some embodiments, I-66 preparations contain no Class 1 or Class 2A elemental impurities. I-66 preparations can be made to meet elemental impurity levels that comply with ICH Q3D limits. In some embodiments, ruthenium levels are about or no more than about 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 25, 30, 35, 40, 45 or 50 ppm. In some embodiments, ruthenium levels are about or no more than about 50 ppm. In some embodiments, ruthenium levels are about or no more than about 40 ppm. In some embodiments, ruthenium levels are about or no more than about 30 ppm. In some embodiments, ruthenium levels are about or no more than about 25 ppm. In some embodiments, ruthenium levels are about or no more than about 20 ppm. In some embodiments, ruthenium levels are about or no more than about 15 ppm. In some embodiments, ruthenium levels are about or no more than about 10 ppm. In some embodiments, palladium levels are about or no more than about 0.01, 0.02, 0.03, 0.04, 0.05, 0.1, 0.2, 0.5, 1.0, 1.5, 2, 5, 10, 15, 20, 25, 30, 40, or 50 ppm. In some embodiments, palladium level is about or no more than about 50 ppm. In some embodiments, palladium level is about or no more than about 20 ppm. In some embodiments, palladium level is about or no more than about 10 ppm. In some embodiments, palladium level is about or no more than about 5 ppm. In some embodiments, palladium level is about or no more than about 2 ppm. In some embodiments, palladium level is about or no more than about 1 ppm. In some embodiments, palladium level is about or no more than about 0.5 ppm. In some embodiments, palladium level is about or no more than about 0.2 ppm. In some embodiments, palladium level is about or no more than about 0.1 ppm. In some embodiments, palladium level is about or no more than about 0.05 ppm. In some embodiments, palladium level is about or no more than about 0.02 ppm.

[0284] Compensial analytical procedures are used to assess I-66 preparations. For example, identity by mass spectrometry, USP <736>; water content, USP <921>; sodium content, USP <1065>; ammonium content, USP <1065>; trifluoroacetic acid content, USP <503.1> / <1065>; acetate content, USP <503>; residual solvents, USP <467>; elemental impurities, USP <232> / <233>; bioburden, USP <61> / <62>; endotoxin, USP <85>; optical rotation, USP <781>; etc.

[0285] Assay and Related Substances Method.

[0286] Operating conditions for identity, purity, and related substances method for I-66 drug substance are provided in Table E-7.

[0287] Table E-7. RP-UHPLC Instrument Conditions for Identity, Purity, and Related Substances Determination for I-66.

Parameter	Condition		
Column:	Waters Acquity Premier Peptide CSH C18, 150 x 2.1 mm, 1.7 μ m, 130 Å, Catalog # 186009489		
Mobile phase:	A: 0.1% trifluoroacetic acid in water B: 0.1% trifluoroacetic acid in acetonitrile		
Diluent	30% acetonitrile/water		
Sample Concentration	1 mg/mL		
Injector Volume:	2 μ L		
Column Temperature	45 °C		
Flow Rate:	0.25 mL/min		
Autosampler Temperature:	5°C $\pm$ 3°C		
Detection	220 nm		
Run Time	50 min		
Gradient Program:	Time (min)	% Mobile Phase A	% Mobile Phase B
	0.0	50	50
	40.0	10	90
	40.1	50	50
	50	50	50

[0288] Peptide Content by Elemental Analysis.

[0289] The peptide content is determined by elemental analysis of the I-66 drug substance using an elemental analyzer. Appropriate quantities of sample are introduced into the elemental analyzer, where it is converted to a gas by combustion at very high temperatures (e.g., 900 °C) in ultrapure oxygen. The nitrogen is converted to NO_x, reduced to N₂, separated in a GC column, and detected using a Thermal Conductivity Detector (TCD). The nitrogen content (%) is used to calculate the peptide content of the sample using the following formula:

$$\text{Peptide content} = \frac{\text{nitrogen content of the sample}}{\text{theoretical nitrogen content}} \times 100\%$$

[0290] Where the theoretical nitrogen content is calculated based on the anhydrous, counterion free substance, i.e., 12.14%, based on the empirical formula for I-66: C₁₀₂H₁₃₄N₁₈O₂₅S₂ (free acid) MW 2076.42.

[0291] Potency Determination

$$\% \text{ Assay} = \frac{[\% \text{ Peptide Purity by RP-HPLC}] \times [\% \text{ Peptide Content}]}{100}$$

[0292] When % peptide purity by RP-HPLC is 93.65 and % peptide content is 90.9, %assay = 85.1%.

[0293] In some embodiments, the present disclosure provides technologies for manufacturing I-66 drug products. Certain preparations are described below as examples.

[0294] In some embodiments, an I-66 drug product is supplied as a 10 mL liquid fill in Type 1 glass

vials with appropriate elastomer stoppers, aluminum seals, and flip-cap. It is labeled for storage at refrigerated conditions (2-8°C). Supportive studies allow for drug administration by IV infusion to be performed at room temperature. I-66 Injection contains 10 mg/mL I-66, 10 mg/mL L-arginine, 33.6 mg/mL PEG-400, and 10 mM sodium phosphate adjusted to pH 8.0. An I-66 drug product formulation is presented below as an example.

[0295] Table E-8. An I-66 drug product formulation.

Component	Amount per mL (mg)	Potential Function
I-66	10	
L-Arginine	10	Solubilizing agent
PEG-400	33.6	Solubilizing agent
Sodium phosphate monobasic anhydrous	1.20	Buffer
0.1 N Sodium hydroxide (NaOH)	Adjust to pH 8.0	pH adjustment
0.1 N Hydrochloric acid (HCl)	Adjust to pH 8.0	pH adjustment
Water for Injection	QS to 1 mL	Solvent

[0296] Various formulations were assessed in order to identify suitable formulations including for stability and solubility. In some embodiments, histidine, phosphate, or Tris buffers in the pH 6.5 to 8.0 range were assessed with Tris and phosphate at pH 7.5 or 8.0 being preferred. In some embodiments, tonicity modifiers sodium chloride or trehalose were assessed. In some embodiments, trehalose was selected based on achieved higher drug solubility. In some embodiments, various amino acid excipients were assessed. In some embodiments, glycine, proline, and arginine were evaluated. In some embodiments, arginine was selected based on higher drug solubility. In some embodiments, to improve solubility, e.g., above about 10 mg/mL, addition of other excipients or cosolvents were assessed. For example, PEG-400, PEG-300, NMP, DMSO, and ethanol were evaluated. In some embodiments, PEG-400 and NMP provided higher solubility increases. In some embodiments, combining PEG-400 (e.g., about 3%) with arginine (e.g., about 10 mg/mL) in phosphate or tris buffer (e.g., about 10 mM) at a suitable pH, e.g., pH 7.5 or 8.0 was able to achieve stable concentrations of I-66 above 25 mg/mL.

[0297] An admixture study was performed to establish the compatibility and stability of I-66 Injection, 10 mg/mL when diluted with 5% Dextrose Injection, USP (D5W) in an IV admixture bag constructed of polyolefin with PVC ports. The dilution concentration was set at the worst case, lowest concentration of 0.24 mg/mL in the D5W bag. The admixture study demonstrated no incompatibilities, degradation, or loss of potency between the D5W solution with either the polyolefin bag or the PVC ports at this concentration. The admixture was shown to be stable for at least 24 hours under ambient room temperature conditions, showing no degradation or loss of potency over the studied time period.

[0298] I-66 Injection, 10 mg/mL is manufactured by adding I-66 Drug Substance to a solution of PEG-400 and L-arginine in sodium phosphate at pH 9.0 ± 0.2 in Water for Injection in the quantities described below. The pH is adjusted with 1N hydrochloric acid to pH 8.0 ± 0.2 and diluted to the target 10 mg/mL

concentration with Water for Injection. The solution is then sterilized by duplicate 0.22 µm filtration and aseptically filled into sterile vials that are stopped and crimped with an aluminum flip-off seal.

[0299] Table E-9. I-66 injection, 10 mg/mL batch formula for 18 L batch size.

Component	Quantity per mL	Batch Quantity	Grade
I-66 Drug Substance	10 mg	180.0 g	Pharmaceutical
L-arginine	10 mg	180.0 g	USP, Ph.Eur.
PEG-400	33.6 mg	604.8 g	NF
Sodium phosphate monobasic, monohydrate	1.2 mg	21.6 g	USP, Ph.Eur.
Sodium hydroxide	pH adjust	pH adjust (40 g ¹)	NF, Ph.Eur., JP
Hydrochloric acid	pH adjust	pH adjust (197 g ²)	NF, Ph.Eur., JP
Water for Injection	Q.S. to 1.0 mL	Q.S. to 18.2 kg	USP, Ph.Eur.

¹Amount needed to make 1000 mL of 1N NaOH solution.

²Amount needed to make 2000 mL of 1N HCl solution.

[0300] The I-66 drug substance stored in the freezer at -20 °C is moved to room temperature to warm before the dispensing. The dispensing of drug substance and all excipients is performed in a downflow booth. Compounding is performed in a Grade C suite. About 60% w/w WFI (13.2 kg ± 0.1 kg) is added to the single use 30 L formulation vessel containing a LevMixer stirrer. To the stirred WFI, PEG-400 (604.8 g ± 1%) is added, followed by L-arginine (180.0 g ± 1%) and sodium phosphate monobasic monohydrate (21.6 g ± 1%) at ambient temperature, and stirring for a minimum of 10 min. between each charge to ensure full dissolution. The pH of the solution is then adjusted to 9.0 ± 0.2 using either 1 N HCl or 1 N NaOH as necessary. I-66 drug substance (180.0 g, correcting for assay) is then charged to the formulation vessel using aliquots of the buffer solution to slurry the drug substance at ambient temperature and in portions. The stirred bulk mixture is pH adjusted to 8.0 ± 0.2 using 1N HCl or 1 N NaOH, at which point a clear solution is obtained. WFI is finally added to a final net weight of 18.2 ± 0.2 kg and the final formulation solution is mixed for NLT 15 min. The final pH of the mixture is measured. A sample is withdrawn for density, appearance, assay, and pre-filtration bioburden analysis.

[0301] In some embodiments, identification of the active pharmaceutical ingredient in the I-66 drug product is performed by the assay and related substances RP-HPLC (Reverse Phase - High Performance Liquid Chromatography) method based on relative retention time.

[0302] Assay and Related Substances by HPLC.

[0303] In some embodiments, operating conditions for the identity, purity, and related substances method for I-66 drug product, e.g., I-66 Injection, 10 mg/mL, are provided below.

[0304] Table E-10. RP-HPLC instrument conditions for identity, purity, assay, and related substances determination for I-66 drug product (e.g., I-66 Injection, 10 mg/mL).

Parameter	Condition
Column	Agilent AdvanceBio Peptide Map 3.0 mm × 100 mm
Mobile Phase	Mobile Phase A: 50 mM Ammonium Phosphate Solution, pH 8.0 in 20% Acetonitrile
	Mobile Phase B: 50 mM Ammonium Phosphate Solution, pH 8.0 in 63%

Parameter	Condition		
	Acetonitrile		
Diluent	50 mM Ammonium Phosphate Solution, pH 8.0 in 40% Acetonitrile		
Sample Concentration	0.5 mg/mL		
Injector Volume	5 μ L		
Column Temperature	45°C		
Flow Rate	0.6 mL/min		
Autosampler Temperature	5°C		
Detection	230 nm		
Run Time	22 min		
Gradient Program	Time (min)	% Mobile Phase A	% Mobile Phase B
	0.00	80	20
	15.00	0	100
	17.00	80	20
	22.00	80	20

[0305] In some embodiments, assay by HPLC is about 90.0-110.0% of label claim. In some embodiments, I-66 HPLC purity is about or at least about 94%, 95%, 96%, 97% or 98%. In some embodiments, pH is about 7.0-9.0.

[0306] In some embodiments, a drug substance or a drug product may contain one or more degradation products. In some embodiments, a drug product may contain higher levels of degradation products compared to a drug product. In some embodiments, based on the amino acid sequence of I-66, anticipated degradation products may be related to degradation of the Asp² and Asp⁵ residues as aspartate and asparagine are amino acids that commonly yield degradation products in peptides. Among other things, the present disclosure provides technologies for I-66 drug substance and drug products that contain no detectable or low levels of degradation products and are suitable for clinical various clinical uses including those described herein.

[0307] Example 2. I-66 is suitable for administration to human subjects for treatment of cancer.

[0308] Extensive studies of I-66 support that it can be utilized for treating human patients. Certain studies are presented below as examples.

[0309] Primary pharmacology studies demonstrated high-affinity binding of I-66 to β -catenin, blockade of β -catenin:TCF4 interactions in biochemical and cell-based assays, inhibition of WNT/ β -catenin downstream transcriptional programs in cancer cell lines and patient-derived xenograft (PDX) tumor models, and anti-tumor effect in PDX models carrying WNT-pathway activating mutations.

[0310] For example, in biochemical assays, direct binding of I-66 to human beta-catenin protein was confirmed by X-ray crystallography. I-66 binds directly to a previously unidentified binding site of beta-catenin that overlaps with the TCF binding site, within the beta-catenin armadillo repeat domain region of the protein. When tested by surface plasmon resonance (SPR), I-66 binds with high affinity to beta-catenin with a dissociation constant (K_d) of 0.5 nM. Competition fluorescence polarization assays (CompFP) demonstrated I-66 blocked beta-catenin:TCF4 interaction with an IC₅₀ <5 nM.

[0311] In cell-based assays, intracellular target engagement was demonstrated by NanoBRET, TCF reporter, quantitative reverse transcription PCR (qPCR) assays, etc. I-66 blocked the interaction of

beta-catenin:TCF4 with an IC_{50} of 1.2 μ M by a NanoBRET assay and inhibited TCF-driven transcriptional activity with an IC_{50} of 0.7 μ M in a TCF luciferase reporter assay. I-66 inhibited endogenous WNT/beta-catenin target gene expression with IC_{50} s in sub or low μ M in cancer cells. In proliferation assays, I-66 displayed IC_{50} in sub or low μ M in cancer cells that carry WPAMs.

[0312] In vivo target engagement was established using RNA sequencing (RNAseq) and protein western blot (WB) analyses. A single dose of I-66 at 20-30 mg/kg significantly down-regulated the WNT/beta-catenin transcriptional pathway in xenograft tumors derived from colorectal cancer (CRC) cell line and CRC patient derived xenograft (PDX) models, which carry WNT pathway activating mutations. I-66 treatment reduced the intratumoral expression of several WNT/beta-catenin target genes in a dose-dependent manner (i.e. CMYC, AXIN2) at both mRNA and protein levels in a CRC PDX model with WNT pathway activating mutations.

[0313] With repeat dosing on a once a week schedule at 20-60 mg/kg dose levels, I-66 led to durable tumor regression or stasis in multiple CRC PDX models and one hepatocellular PDX model harboring WNT pathway activating mutations. No anti-tumor effects were observed in tested tumor xenograft models that do not carry known activating mutations of the WNT pathway. AXIN2 and C-MYC can both be relevant pharmacodynamic biomarkers since inhibition of these biomarkers is on-target, dose dependent, correlated with efficacy and in the case of C-MYC, without the intention to be bound by particular theory, likely to be relevant for the anti-tumor activity of I-66 downstream of beta-catenin inhibition. Significant inhibition of AXIN2 and C-MYC is observed in tumors starting at 20 mg/kg and maximal inhibition is achieved at 60 mg/kg.

[0314] Many studies, including secondary pharmacology, safety pharmacology, pharmacokinetics, toxicology, etc., were performed, confirming that I-66 can be administered as described herein for treating cancer.

[0315] Nonclinical pharmacology programs included an assessment of the potential of I-66 to modulate binding and activity of many off-target receptors, enzymes, ion channels, and transporters. Safety pharmacology studies were conducted with animals including monkeys. PK of I-66 was investigated in animals including BALB/c mice, Sprague Dawley rats, and cynomolgus monkeys to assess absorption, distribution, metabolism, and excretion. Both single- and multiple-dose intravenous (IV) administration were evaluated. In addition, in vitro metabolism studies were conducted with animals including rat, dog, monkey, and human liver microsomes and hepatocytes.

[0316] The toxicity profile of I-66 was evaluated including in vitro in a bacterial reverse mutation assay and in a Chinese Hamster Ovary (CHO) cell micronucleus assay, and in vivo in Sprague Dawley rats and cynomolgus monkeys. In vivo studies included two non-GLP repeat-dose studies in rats (one with Q4D dosing and one with Q7D dosing), one non-GLP study in monkeys with an escalating dose phase and a repeat-dose phase, and two 5-week repeat-dose pivotal GLP compliant studies: one in rats and one in monkeys.

[0317] No modulation of ion channel current up to the highest concentration tested was observed. ECG tracings in monkeys did not show any test article related abnormalities in cardiac rhythm or conduction when $C_{\max}$ values were many-fold high over the projected human $C_{\max}$ value ranges.

[0318] Pharmacokinetic and toxicokinetic profiles confirmed I-66 can be administered to human subjects for cancer treatment as described herein.

[0319] I-66 exhibited high protein binding in human (2.1 to 4.1% unbound) and nonclinical species plasma, without concentration-dependent changes. I-66 bound preferentially to human serum albumin in comparison to alpha1-acid glycoprotein. I-66 showed low partition to red blood cells across species.

[0320] I-66 is stable for use as described herein. For example, I-66 is stable in plasma, whole blood, and liver microsomes of at least BALB/c mice, SD rat, cynomolgus monkey and human. It is also stable in liver S9 fraction derived from rats and human. I-66 was the predominant component following incubation in hepatocytes from mice, rats, monkeys and humans for up to 24 hours. There was no human unique metabolite in hepatocytes; rat and monkeys were deemed appropriate toxicology species based on the similarity of in vitro metabolism to human. I-66 was also assessed for liver microsome. In some embodiments, drugs that are substrates of the relevant CYPs (1A2, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4) that have a narrow therapeutic index may not be co-administered with I-66. In some embodiments, drugs that are MDR1 or BCRP substrates and have narrow therapeutic indices (e.g., digoxin and methotrexate) may not be co-administered with I-66. In some embodiments, caution should be exercised when co-administering medications that are substrates of OATP1B1 and OATP1B3 with I-66. In some embodiments, medications that are substrates of OATP1B1 and OATP1B3 are not co-administered with I-66.

[0321] I-66 is not a substrate of the uptake solute carrier (SLC) transporters MATE1, MATE2-K, OAT1, OAT3, OCT1, OCT2 and OATP1B1, and is marginally a substrate of OATP1B3.

[0322] Toxicology of I-66 was assessed in various assays including animals such as rat and monkey. Based on the results of the nonclinical toxicology programs, I-66 has an acceptable safety profile for use as described herein.

[0323] Various safety pharmacology, toxicology, and toxicokinetics (TK) studies were conducted in accordance with guidelines issued by the ICH and GLP.

[0324] I-66 has been compared to various other stapled peptide agents that can bind to beta-catenin in various assays including those described herein. In various cases, I-66 demonstrated superior properties and activities, including higher potency for inhibiting beta-catenin : TCF interactions, modulating transcription, inhibiting cancer cell proliferation, etc.

[0325] Among other things, results described herein demonstrate an acceptable benefit/risk profile for administering I-66 as an anticancer therapeutic for patients with known or suspected WPAM.

[0326] Example 3. Clinical trial designs to demonstrate safety and efficacy of I-66.

[0327] Among other things, the present disclosure provides technologies including clinical trials for assessing and confirming safety and efficacy of I-66. Useful examples are described below.

[0328] In some embodiments, I-66 is assessed to confirm for treatment of advanced and/or metastatic solid tumors. I-66 Injection, e.g., 10 mg/mL, is supplied as a sterile, aseptically processed solution labeled for storage at 2-8°C in single-use vials for intravenous (IV) administration. The initial dosing schedule is weekly (QW) in 28-day cycles and can continue until disease progression or the occurrence of unmanageable toxicity. A study scheme is provided in Figure 13 as an example. For administration, a solution (e.g., a liquid composition of I-66 of 10 mg/mL or higher as described herein) comprising the amount of I-66 to be administered (e.g., about 18, 36, 72, 144, 240, 360, 480, 600, etc. mg/m²) is diluted with a pharmaceutically acceptable solution to produce a solution for administration that is of a set volume. In some cases, a diluent is 5% Dextrose Injection, USP. In some cases, a set volume of solution for administration is 250 mL. A preparation procedure, which dilutes I-66 in 5% Dextrose Injection, USP, to a total volume of 250 mL for intravenous infusion, is described below as an example:

obtain the number of vials required (one vial contains 10 mL (100 mg) of I-66 at a concentration of 10 mg/mL);

visually inspect the vial(s) for particulate matter and discoloration prior to administration. I-66 Injection, 10 mg/mL, is provided as a clear to slightly opalescent, colorless to slightly tinted solution, essentially free of visible particulates. Do not use if particles are present in the solution;

remove a volume of 5% Dextrose Injection from the infusion bag/container that is equal to the calculated and required volume of I-66 Injection, 10 mg/mL, plus the estimated volume of overfill, if applicable. In some embodiments, an infusion bags/containers is DEHP-free, PVC-free, and natural rubber latex-free, and made of either polyethylene (PE) or polypropylene (PP), polyamide (PA), or a combination thereof;

withdraw the necessary amount of I-66 Injection, 10 mg/mL and dilute by adding to the infusion bag/container containing 5% Dextrose Injection. When applicable, the same syringe may be used to withdraw the full volume from multiple vials. The total volume of I-66 solution in the infusion bag/container is about 250 mL; and

gently invert the infusion bag, e.g., about 10 times, to mix the diluted solution.

[0329] I-66 can be administered QW intravenously, e.g., on Days 1, 8, 15 and 22 of each 28-day cycle, over approximately 60-75 minutes. For doses of about 500 mg/m² or more, infusion time can be increased to about 75 minutes. After administration of I-66, an injection line can be flushed, e.g., with about 25 mL or more of 5% Dextrose Injection.

[0330] A useful protocol for calculating the amount of I-66 for administration to a patient is described below as an example:

calculate the Body Surface Area (BSA) of a patient using the Mosteller formula as below:

$$BSA (m^2) = \sqrt{\frac{Height (cm) \times Weight (Kg)}{3,600}}$$

;

calculate the volume of I-66 Injection required (using 10 mg/mL as an example):

$$\frac{\text{Dose (mg/m}^2\text{)} \times \text{BSA (m}^2\text{)}}{10\text{mg/mL}} = \text{I-66 Injection, 10 mg/mL (mL)}$$

[0331] For example, a patient with a calculated BSA of 2.0 m² at 36 mg/m² requires about 7 mL of I-66 Injection, 10 mg/mL:

$$36\text{mg/m}^2 \times 2.0\text{m}^2 \text{ divided by } 10 \text{ mg/mL} = 7.2 \text{ mL (if round to whole number, 7 mL)}$$

The about 7 mL I-66 10 mg/mL can be diluted using 5% Dextrose Injection, USP to about 250 mL for IV as described herein.

[0332] Certain examples of I-66 diluted compositions for administration are described below as examples.

Table E-11. Example diluted solutions for 250 mL administration volume.

Dose (mg/m ²)	BSA (m ²)					
	Component	1.3	1.5	1.7	1.9	2.1
36	I-66 (mg/mL)	0.2	0.2	0.2	0.3	0.3
	L-arginine (mg/mL)	0.2	0.2	0.2	0.3	0.3
	PEG-400	0.6	0.7	0.8	0.9	1.0
	Sodium phosphate monobasic, monohydrate (mg/mL)	0.1	0.1	0.1	0.1	0.1
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.05	0.05	0.05	0.05	0.05
72	I-66 (mg/mL)	0.4	0.4	0.5	0.5	0.6
	L-arginine (mg/mL)	0.4	0.4	0.5	0.5	0.6
	PEG-400	1.3	1.5	1.6	1.8	2.0
	Sodium phosphate monobasic, monohydrate (mg/mL)	0.2	0.2	0.2	0.2	0.2
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.05	0.05	0.05	0.05	0.05
144	I-66 (mg/mL)	0.7	0.9	1.0	1.1	1.2
	L-arginine (mg/mL)	0.7	0.9	1.0	1.1	1.2
	PEG-400	2.5	2.9	3.3	3.7	4.1
	Sodium phosphate monobasic, monohydrate (mg/mL)	0.3	0.3	0.4	0.4	0.5
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.05	0.05	0.05	0.04	0.04
240	I-66 (mg/mL)	1.2	1.4	1.6	1.8	2.0
	L-arginine (mg/mL)	1.2	1.4	1.6	1.8	2.0

Dose (mg/m ²)	BSA (m ²)					
	Component	1.3	1.5	1.7	1.9	2.1
	PEG-400	4.2	4.8	5.5	6.1	6.8
	Sodium phosphate monobasic, monohydrate (mg/mL)	0.5	0.6	0.7	0.7	0.8
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.04	0.04	0.04	0.04	0.04
360	I-66 (mg/mL)	1.9	2.2	2.4	2.7	3.0
	L-arginine (mg/mL)	1.9	2.2	2.4	2.7	3.0
	PEG-400	6.3	7.3	8.2	9.2	10.2
	Sodium phosphate monobasic, monohydrate (mg/mL)	0.8	0.9	1.0	1.1	1.2
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.04	0.04	0.04	0.04	0.03
480	I-66 (mg/mL)	2.5	2.9	3.3	3.6	4.0
	L-arginine (mg/mL)	2.5	2.9	3.3	3.6	4.0
	PEG-400	8.4	9.7	11.0	12.3	13.5
	Sodium phosphate monobasic, monohydrate (mg/mL)	1.0	1.2	1.3	1.5	1.6
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.04	0.04	0.03	0.03	0.03
600	I-66 (mg/mL)	3.1	3.6	4.1	4.6	5.0
	L-arginine (mg/mL)	3.1	3.6	4.1	4.6	5.0
	PEG-400	10.5	12.1	13.7	15.3	16.9
	Sodium phosphate monobasic, monohydrate (mg/mL)	1.3	1.5	1.6	1.8	2.0
	Sodium hydroxide*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Hydrochloric acid*	pH adjust	pH adjust	pH adjust	pH adjust	pH adjust
	Dextrose (mg/mL)	0.03	0.03	0.03	0.03	0.02

*pH adjusted to about 8.0 before dilution with 5% Dextrose Injection, USP.

[0333] In some embodiments, dose escalation comprises 2 parallel components: Dose Escalation (Phase 1a) and Pharmacodynamic (PD) Biomarker Cohorts (Phase 1b). In some cases, in Part 1a, the safety, tolerability, and PK of escalating dose levels of I-66 is evaluated sequentially in a standard 3+3 design. Participants with microsatellite stable (MSS) CRC or any solid tumor with a documented WPAM are enrolled. In parallel with Dose Escalation, additional participants with MSS CRC are enrolled in PD Biomarker Cohorts (Part 1b). Paired tumor biopsies are performed predose and while on-study, and evaluated for markers of pharmacodynamic effects of I-66, including Myc expression.

[0334] Upon selection of a recommended Phase 2 dose (RP2D), Dose Expansion (Phase 2) evaluate the antitumor activity of I-66 in defined patient populations that are expected or documented to have a WPAM, including MSS CRC, gastric/gastroesophageal (GEJ) cancer, and non-small cell lung cancer (NSCLC). Participants with CRC may not be required to be screened for WPAMs since the majority of these patients are known to have pathway activation but participants with gastric/GEJ cancer or NSCLC may be required to have a WPAM to be enrolled, based on local testing.

[0335] Dose Escalation (Parts 1a and 1b): In some cases, I-66 is administered at escalating doses ranging from 36 to 600 mg/m². I-66 is administered over 1 hour via intravenous (IV) infusion on Days 1, 8, 15, and 22 of each 28-day cycle. For any dose above 500 mg/m², administration time is increased from 1 hour to at least 75 minutes. Additional dosing regimens and IV infusion durations may be explored in dose escalation Part 1. In some cases, in Part 1a, a standard 3+3 dose escalation of single-agent I-66 in participants with solid tumor known to harbor a WPAM or microsatellite stable (MSS) CRC (irrespective of WPAM status) is evaluated. Typically, a minimum of 3 eligible participants are assigned to each dose level during the dose-escalation period. Participants may continue treatment for additional cycles until disease progression, unacceptable toxicity, or treatment discontinuation for another reason occurs. PK analysis can be performed in near real-time on samples collected during dose escalation. In some cases, in Part 1b, additional cohorts of up to 6 participants may be dosed to evaluate the pharmacodynamic effects of I-66 on MSS CRC tumors. Part 1b of the study is not opened until a cohort in Part 1a achieves sufficient PK exposure of I-66 that pharmacodynamic modulation of the Wnt pathway is considered likely. These cohorts are opened at dose levels that have previously been considered tolerable in Part 1a of the study. In some cases, participants have fresh core needle biopsies. The first biopsy is performed prior to dosing (i.e., during screening) and the second biopsy is performed within 5 days following the dose on either Cycle 1 Day 15 (C1D15), Cycle 1 Day 22 (C1D22), or Cycle 2 Day 1 (C2D1).

[0336] Dose Expansion (Part 2): I-66 is administered at the RP2D via IV infusion at a frequency that has been considered safe and tolerable during Part 1 of the study. Additional dosing regimens may be explored in dose expansion Part 2. The following cohorts of participants may be enrolled to receive single-agent I-66 at the RP2D:

- Cohort 2a: Participants with advanced or metastatic MSS CRC. Enrollment to this cohort follows a Simon 2-stage design where the observed clinical activity in the first 12 participants determines whether additional participants are enrolled for a total of about 37 participants.
- Cohort 2b: Participants with advanced or metastatic NSCLC with a documented WPAM in APC or beta-catenin, e.g., as identified by next-generation sequencing (NGS) assay performed on DNA extracted from tumor tissue or plasma (i.e., circulating tumor DNA [ctDNA]). Enrollment to this cohort follows a Simon 2-stage design where the observed clinical activity in the first 12 participants determines whether additional participants are enrolled for a total of about 37 participants.

- Cohort 2c: Participants with advanced or metastatic gastric/gastroesophageal junction (GEJ) cancer with a documented WPAM in APC or beta-catenin, e.g., as identified by a tumor tissue- or ctDNA-based NGS assay. Enrollment to this cohort follows a Simon 2-stage design where the observed clinical activity in the first 12 participants determines whether additional participants are enrolled for a total of about 37 participants.
- Cohort 2d: Initially, up to 30 participants with solid tumors harboring at least one documented WPAM in any of the predefined Wnt-pathway genes are enrolled. This also includes CRC, NSCLC, and gastric/GEJ cancers with a WPAM but are ineligible for Cohorts 2a, 2b, or 2c. If objective responses are seen in this cohort, enrollment of that patient population may be expanded in a single-arm Part 2 cohort (to be added via a formal protocol amendment). Predefined Wnt pathway activating mutations are *APC* loss of function mutations (LOF, including frameshift, nonsense, or splice site), *CTNNB1* gain of function mutations (GOF, including missense, in-frame insertion, or deletion at codons 32-37, 41, and 45), *RNF43* LOF mutations, and *RSPO2* and *RSPO3* fusions. Other genomic alterations of the above genes and in other Wnt-pathway genes, such as *AMER1*, *AXIN1*, *AXIN2*, *BCL9*, *CSNK1A1*, *GSK3B*, *LRP5*, *LRP6*, *LGR5*, *TCF7L2*, and *WIF1*, may be considered.

[0337] For Cohorts 2a, 2b, and 2c, the decision to continue enrollment in Stage 2 can be made once the target number of confirmed objective responses has been achieved. If the target number of objective responses has not been achieved within 24 weeks of enrolling the last Stage 1 participant, the totality of the clinical data will be reviewed, and a decision made regarding the cohort.

[0338] An estimated duration of study participation is up to 28 days for screening, then continuous treatment in consecutive 28-day cycles for as long as participants are receiving benefit, are tolerating the regimen, and do not meet any criteria for discontinuation of study treatment, and 30 days after the last dose for safety follow-up. Study participation, including post-treatment follow-up, is expected to average approximately 6 months per participant. In some cases, a cycle is a 21-day cycle.

[0339] In some embodiments, primary purpose is treatment. In some embodiments, allocation is non-randomized. In some embodiments, about 200 participants are enrolled. In some embodiments, there are no masking. In some embodiments, interventional study model is sequential assignment. In some embodiments, there are 6 arms, for example:

Arms	Assigned Interventions
Experimental: Part 1a Solid Tumors with Any WNT-Pathway Activating Mutation (WPAM) or Microsatellite Stable (MSS) Colorectal Cancer (Irrespective of WPAM Status)	Drug: I-66 I-66 will be administered IV once weekly at assigned doses in continuous cycles of 28 days
Experimental: Part 1b Microsatellite Stable Colorectal Cancer	Drug: I-66 I-66 will be administered IV once weekly at assigned doses in continuous cycles of 28 days

Arms	Assigned Interventions
Experimental: Cohort 2a Microsatellite Stable Colorectal Cancer	Drug: I-66 I-66 will be administered IV once weekly at the RP2D dose in continuous cycles of 28 days
Experimental: Cohort 2b Non-Small Cell Lung Cancer with a WNT-Pathway Activating Mutation (WPAM) in Adenomatous Polyposis Coli (APC) or Beta-Catenin	Drug: I-66 I-66 will be administered IV once weekly at the RP2D dose in continuous cycles of 28 days
Experimental: Cohort 2c Gastric Cancer/Gastroesophageal Junction Carcinoma (GEJ) with a WNT-Pathway Activating Mutation (WPAM) in Adenomatous Polyposis Coli (APC) or Beta-Catenin	Drug: I-66 I-66 will be administered IV once weekly at the RP2D dose in continuous cycles of 28 days
Experimental: Cohort 2d Solid Tumors with Any WNT-Pathway Activating Mutation (WPAM)	Drug: I-66 I-66 will be administered IV once weekly at the RP2D dose in continuous cycles of 28 days

In some embodiments, about 240 participants are enrolled. In some embodiments, a study design enrolls about 245 participants. In some embodiments, a cycle is a 21-cycle. In some embodiments, arms and interventions in a study is presented below as an example:

Arms	Intervention/treatment
Experimental: Part 1a Solid Tumors with Any WNT-Pathway Activating Mutations (WPAMs) or Microsatellite Stable (MSS)Colorectal Cancer (Irrespective of WPAM Status)	Drug: I-66 I-66 will be administered IV at assigned doses in continuous cycles of 21 or 28 days
Experimental: Part 1b Microsatellite Stable Colorectal Cancer (Irrespective of WPAM Status)	Drug: I-66 I-66 will be administered IV at assigned doses in continuous cycles of 21 or 28 days
Experimental: Cohort 2a Microsatellite Stable Colorectal Cancer (Irrespective of WPAM Status)	Drug: I-66 I-66 will be administered IV once weekly at the preliminary RP2D dose in continuous cycles of 21 or 28days
Experimental: Cohort 2b Non-Small Cell Lung Cancer with a WNT-Pathway Activating Mutation(WPAM) in Adenomatous Polyposis Coli (APC) or Beta-Catenin	Drug: I-66 I-66 will be administered IV once weekly at the preliminary RP2D dose in continuous cycles of 21 or 28days
Experimental: Cohort 2c Gastric Cancer/Gastroesophageal Junction Carcinoma (GEJ) with a WNT-Pathway Activating Mutation (WPAM) in Adenomatous Polyposis Coli (APC) or Beta-Catenin	Drug: I-66 I-66 will be administered IV once weekly at the preliminary RP2D dose in continuous cycles of 21 or 28days
Experimental: Cohort 2d Solid Tumors with Any WNT-Pathway	Drug: I-66 I-66 will be administered IV once weekly at

Arms	Intervention/treatment
Activating Mutations (WPAMs)	the preliminary RP2D dose in continuous cycles of 21 or 28days

[0340] In some embodiments, I-66 is administered IV weekly. In some embodiments, I-66 is administered about every two weeks. In some embodiments, I-66 is administered about every three weeks.

[0341] Useful outcome measures include:

Primary Outcome Measure:

1. During dose escalation and dose expansion measure incidence and severity of treatment emergent adverse events by CTCAE v5.0

Number and severity of treatment emergent adverse events as assessed by CTCAE v5.0

[Time Frame: Through study completion, an average of 4 months]

2. During dose escalation characterize dose-limiting toxicities (DLTs)

Incidence of DLTs

[Time Frame: 1 treatment cycle (e.g., 28 days)]

3. During dose expansion describe the Overall Response Rate using RECIST v1.1

The rate of objective responses (Partial & Complete) using RECIST v1.1

[Time Frame: Every 56 days until study completion, approximately 4 months on average]

Secondary Outcome Measure:

4. Pharmacokinetic profile

Study drug exposure over time

Plasma concentration (C_{max}) of I-66

Time to achieve C_{max} (T_{max}) of I-66 in plasma

Area under the plasma concentration-time curve (AUC) of I-66

Clearance (CL) of I-66 from the plasma

Terminal half-life (t_{1/2}) of I-66 in plasma

[Time Frame: During first 2 cycle (e.g., 56 days)]

5. During dose escalation select the (preliminary) recommended Phase 2 dose and dosing schedule of study drug

Rate of Dose Limiting Toxicities (DLTs) across dose levels

[Time Frame: During Cycle 1 (e.g., 28 days)]

6. During dose escalation Part 1b to evaluate the pharmacodynamic activity in tumors

Change in tumor Myc expression (on-study compared to baseline)

[Time Frame: During first 2 cycles (e.g., 56 days)]

7. During dose escalation and expansion to describe Best Overall Response Rate using RECIST v1.1

Best response to treatment using RECIST v1.1

[Time Frame: Every 56 days until study completion, approximately 4 months on average]

8. During dose escalation and expansion to describe Duration of Response using RECIST v1.1

Time from initial objective response (partial response (PR) or complete response (CR)) to disease progression

[Time Frame: Every 56 days until study completion, approximately 4 months on average]

9. During dose expansion describe Progression Free Survival

Progression Free Survival (PFS) using RECIST v1.1

[Time Frame: From date of randomization until the date of first disease progression, an average of 4 months (in some embodiments, the first documented disease progression, or death, an average of 4 months and/or assessed for up to 18 months).]

[0342] In some embodiments, the following eligibility criteria are utilized:

Ages Eligible for Study: 18 Years and older

Sex: All

Gender Based: No

Accepts Healthy Volunteers: No

Criteria:

Inclusion Criteria:

- Eastern Cooperative Oncology Group (ECOG) performance status of 0 or 1.
- Adequate organ and marrow function.

Additional Inclusion Criteria for Dose Escalation Cohorts (Part 1a):

- Diagnosis of treatment-refractory advanced/metastatic solid tumor that is either non-MSI-H or non-dMMR colorectal cancer (CRC) or any other solid tumor with documented WNT- pathway activating mutations (WPAMs).

Additional Inclusion Criteria for Dose Escalation Cohorts (Part 1b):

- Diagnosis of treatment-refractory advanced/metastatic non-MSI-H or non-dMMR CRC
- At least one lesion that is suitable for core needle biopsy.

Additional Inclusion Criteria for Dose Expansion Cohort (Cohort 2a): Colorectal Cancer (CRC) Cohort

- Diagnosis of treatment-refractory advanced/metastatic non-MSI-H or non-dMMR CRC.

Additional Inclusion Criteria for Dose Expansion Cohort (Cohort 2b): Non-Small Cell Lung Cancer (NSCLC) Cohort

- Diagnosis of treatment-refractory advanced/metastatic NSCLC with documented WPAMs in adenomatous polyposis coli (APC) or Beta-catenin.

Additional Inclusion Criteria for Dose Expansion Cohort (Cohort 2c): Gastric/Gastroesophageal junction (GEJ) Cohort

- Diagnosis of treatment-refractory advanced/metastatic gastric/GEJ cancer with documented WPAMs in adenomatous polyposis coli (APC) or Beta-catenin.

Additional Inclusion Criteria for Dose Expansion Cohort (Cohort 2d): Tumor Agnostic Cohort

- Diagnosis of treatment-refractory advanced/metastatic solid tumor with documented WPAMs.

Exclusion Criteria:

- Known history of bone metastasis.
- Evidence of vertebral compression fracture or non-traumatic bone fracture within the past 12 months and who are not receiving antiresorptive therapy.
- Osteoporosis, which is defined as a T-score of <-2.5 at the lumbar spine (L1 - L4), left (or right) femoral neck or left (or right) total hip as determined by DXA scan, or a FRAX 10-year probability of hip fracture $\geq 3\%$ or a 10-year probability of major osteoporosis-related fracture $\geq 20\%$, based on the US-adapted WHO algorithm for postmenopausal women and men ≥ 50 years of age.
- Inflammatory bowel disease (i.e., ulcerative colitis or Crohn's disease) that is recently active or requires therapy currently.
- Unstable/inadequate cardiac function
- Has known meningeal carcinomatosis, leptomeningeal carcinomatosis, spinal cord compression, or symptomatic or unstable brain metastases.
- Pregnant, lactating, or planning to become pregnant.

[0343] Certain useful technologies are described in a clinical trial with a ClinicalTrials.gov ID NCT05919264. In some embodiments, a clinical trial for demonstrating safety, efficacy, etc. of provided technologies is or comprises or is similar to a clinical trial with a ClinicalTrials.gov ID NCT05919264.

[0344] In some embodiments, a dose is about 18 mg/m². In some embodiments, a dose is about 36 mg/m². In some embodiments, a dose is about 72 mg/m². In some embodiments, a dose is about 144 mg/m². In some embodiments, a dose is about 240 mg/m². In some embodiments, a dose is about 360 mg/m². In some embodiments, a dose is about 480 mg/m². In some embodiments, a dose is about 600 mg/m². In some embodiments, a patient may start with a lower dose. In some embodiments, a patient may start with a higher dose. In some embodiments, there are one or more loading doses. In some embodiments, there are no loading doses. In some embodiments, doses are administered about weekly. In some embodiments, safety and/or efficacy are to be observed for one or more dose levels in view of existing study results. Among other things, provided technologies evaluate and demonstrate safety, tolerability, pharmacodynamics (PD), pharmacokinetics (PK), antitumor activity, etc. of I-66, e.g., in patients with advanced or metastatic solid tumors. I-66 has been administered to a number of patients according to several regimens described above and has demonstrated profiles suitable for continued clinical trials (see, e.g., NCT05919264).

[0345] While various embodiments have been described and illustrated herein, those of ordinary skill in the art will readily envision a variety of other means and/or structures for performing the functions and/or obtaining the results and/or one or more of the advantages described in the present disclosure, and each of such variations and/or modifications is deemed to be included. More generally, those skilled in the art will readily appreciate that all parameters, dimensions, materials, and configurations described herein are meant to

be example and that the actual parameters, dimensions, materials, and/or configurations will depend upon the specific application or applications for which the teachings of the present disclosure is/are used. Those skilled in the art will recognize, or be able to ascertain using no more than routine experimentation, many equivalents to the specific embodiments of the disclosure described in the present disclosure. It is, therefore, to be understood that the foregoing embodiments are presented by way of example only and that, provided technologies, including those to be claimed, may be practiced otherwise than as specifically described and claimed. In addition, any combination of two or more features, systems, articles, materials, kits, and/or methods, if such features, systems, articles, materials, kits, and/or methods are not mutually inconsistent, is included within the scope of the present disclosure.

CLAIMS

1. A composition, comprising:
I-66;
a buffer agent;
optionally a pH adjustment agent; and
a solvent.
2. The composition of claim 1, wherein the composition comprises a PEG.
3. The composition of any one of the preceding claims, further comprising an amino acid.
4. The composition of any one of the preceding claims, further comprising a surfactant.
5. The composition of claim 1, further comprising a solubilizing agent.
6. The composition of any one of claims 3-5, further comprising a solubilizing agent.
7. The composition of claim 6, wherein the solubilizing agent is a PEG or a surfactant.
8. The composition of any one of the preceding claims, wherein the composition is a liquid composition.
9. The composition of any one of the preceding claims, comprising an I-66 pharmaceutically acceptable salt form.
10. The composition of any one of the preceding claims, comprising an I-66 sodium salt form.
11. The composition of any one of the preceding claims, wherein I-66 is Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and wherein:

¹³C NMR of I-66 in MeOD at 25°C, comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58, and I-66 has a purity of about or at least about 90%; and/or

¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84,

1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or ¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

12. The composition of any one of the preceding claims, wherein identity of I-66 is confirmed by comparison with a reference standard, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and/or

when characterized by ¹H NMR in MeOD at 25°C, its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H),

2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

13. The composition of claim 12, wherein the comparison is or comprises utilizing HPLC, optionally wherein the HPLC condition is selected from those in Example 1, as described in Table E-7 or as described in Table E-10.

14. The composition of any one of claims 12-13, wherein the comparison is or comprises utilizing NMR, optionally wherein the NMR is ¹H NMR, ¹³C NMR, and/or two- or multi-dimensional NMR.

15. The composition of any one of the preceding claims, wherein the concentration of I-66 is about 1-25 mg/mL, preferably wherein the concentration of I-66 is about 10 mg/mL.

16. The composition of any one of claims 3-15, wherein the amino acid is a basic amino acid.

17. The composition of any one of claims 3-16, wherein the amino acid is arginine.

18. The composition of any one of claims 3-17, wherein the concentration of the amino acid is about 1-20 mg/mL, preferably wherein the concentration of the amino acid is about 10 mg/mL.

19. The composition of any one of the preceding claims, comprising a PEG-400.

20. The composition of any one of the preceding claims, wherein the concentration of the PEG is about 1%-5% w/v or 1%-5% v/v, preferably wherein the concentration of PEG is about 3.4% w/v, about 3% w/v, about 3.4% v/v, or about 3% v/v.

21. The composition of any one of the preceding claims, comprising a nonionic surfactant.

22. The composition of any one of the preceding claims, comprising Tween 80.

23. The composition of any one of the preceding claims, wherein the concentration of the surfactant is about 0.5%-5% w/v or about 0.5%-5% v/v, preferably wherein the concentration of the surfactant is about 1% w/v or 1% v/v.

24. The composition of any one of the preceding claims, wherein the buffering agent is a phosphate salt,

preferably wherein the buffering agent is sodium phosphate monobasic.

25. The composition of any one of claims 1-23, wherein the buffering agent is Tris.
26. The composition of any one of the preceding claims, wherein the concentration of the buffering agent is about 10 mM.
27. The composition of any one of the preceding claims, comprising a pH adjusting agent.
28. The composition of claim 27, wherein the pH adjusting agent is NaOH or HCl.
29. The composition of any one of the preceding claims, wherein the solvent is water.
30. The composition of any one of the preceding claims, comprising I-66, arginine, PEG-400, sodium phosphate buffer, and water.
31. The composition of any one of the preceding claims, comprising or consisting of I-66, arginine, PEG-400, sodium phosphate, and water, and having a pH of about 7.0-9.0 adjusted using NaOH and HCl.
32. The composition of any one of the preceding claims, consisting of I-66, arginine, PEG-400, sodium phosphate, and water, and having a pH of about 7.0-9.0 adjusted using NaOH or HCl.
33. The composition of any one of the preceding claims, wherein the pH of the composition is about 7.0-9.0 or about 8.0.
34. A composition, comprising:
 - I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
 - a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% w/v;
 - a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
 - optionally a pH adjustment agent; and
 - water;
 - wherein the pH of the composition is about 7.0-9.0; ora composition, comprising:
 - I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - arginine, wherein the concentration of arginine is about 1-20 mg/mL;
 - PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
 - a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
 - optionally a pH adjustment agent; and
 - water;
 - wherein the pH of the composition is about 7.0-9.0; ora composition, comprising:
 - I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
 - arginine, wherein the concentration of arginine is about 1-20 mg/mL;
 - PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;
 - sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

1-100 mM;

optionally a pH adjustment agent; and

water;

wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

arginine, wherein the concentration of arginine is about 1-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

1-50 mM;

optionally sodium hydroxide;

optionally hydrochloric acid; and

water;

wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

arginine, wherein the concentration of arginine is about 1-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

1-50 mM;

optionally sodium hydroxide;

optionally hydrochloric acid; and

water;

wherein the pH of the composition is about 7.0-9.0; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

arginine, wherein the concentration of arginine is about 5-15 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

1-50 mM;

optionally sodium hydroxide;

optionally hydrochloric acid; and

water;

wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-100 mM;
optionally a pH adjustment agent; and
water;
wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 1-50 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0.

35. The composition of claim 34, wherein the concentration of PEG-400 is about 3% w/v.

36. The composition of any one of claims 34-35, wherein the concentration of I-66 is about or at least about 10 mg/mL, or about 10-25 mg/mL.

37. The composition of any one of claims 34-36, wherein the concentration of the amino acid or arginine is about 10 mg/mL.

38. The composition of any one of claims 34-37, wherein the concentration of PEG-400 is about 3% v/v.

39. The composition of any one of claims 34-38, wherein the concentration of the buffer agent or sodium phosphate monobasic is about 5-20 mM, or is about 10 mM.

40. A composition, comprising:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;

- optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;

- PEG-400, wherein the concentration of PEG-400 is about 2%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% w/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid; and
water;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 1-20 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.5%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:

- I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.5%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 1-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 1-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 5-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-20 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 2%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:
I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3%-5% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 5-15 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:

I-66, wherein the concentration of I-66 is about 10-25 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, comprising:

I-66, wherein the concentration of I-66 is about 10 mg/mL;
arginine, wherein the concentration of arginine is about 10 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
a buffer;
wherein the pH of the composition is about 7.0-9.0; or
a composition, consisting of:

- I-66, wherein the concentration of I-66 is about 10 mg/mL;
 arginine, wherein the concentration of arginine is about 10 mg/mL;
 PEG-400, wherein the concentration of PEG-400 is about 3% v/v; and
 a buffer;
 wherein the pH of the composition is about 7.0-9.0.
41. The composition of claim 40, wherein the concentration of I-66 is about 10 mg/mL.
42. The composition of claim 40, wherein the concentration of I-66 is about or at least about 10 mg/mL or is about 10-25 mg/mL.
43. The composition of any one of claims 40-42, wherein the system comprises a buffer and the buffer is a phosphate buffer, optionally a sodium phosphate buffer.
44. The composition of any one of claims 40-42, wherein the system comprises a buffer and the buffer is a Tris buffer.
45. The composition of any one of claims 40-46, wherein the system comprises a buffer and the concentration of the buffer is about 5-100 mM, optionally about 10 mM.
46. A composition, comprising:
 10 mg/mL I-66;
 10 mg/mL arginine;
 33.6 mg/mL PEG-400; and
 10 mM sodium phosphate;
 wherein the pH of the composition is about 8.0; or
 a composition, consisting of:
 10 mg/mL I-66;
 10 mg/mL arginine;
 33.6 mg/mL PEG-400; and
 10 mM sodium phosphate;
 wherein the pH of the composition is about 8.0.
47. The composition of any one of claims 34-46 wherein I-66 is Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and wherein:
¹³C NMR of I-66 in MeOD at 25°C, comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13,

139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58, and I-66 has a purity of about or at least about 90%; and/or

¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise, or ¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

48. The composition of any one of claims 34-47, wherein identity of I-66 is confirmed by comparison with a reference standard, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ¹H NMR in MeOD at 25°C, its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60,

1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

49. The composition of claim 48, wherein the comparison is or comprises utilizing HPLC, optionally wherein the HPLC condition is selected from those in Example 1, as described in Table E-7 or as described in Table E-10.

50. The composition of any one of claims 48-49, wherein the comparison is or comprises utilizing NMR, optionally wherein the NMR is ¹H NMR, ¹³C NMR, and/or two- or multi-dimensional NMR.

51. The composition of any one of the preceding claim, further comprising a pharmaceutically acceptable carrier.

52. The composition of any one of the preceding claims, further comprising a sugar, optionally wherein the sugar is dextrose.

53. A composition, prepared by diluting a composition of any one of claims 1-52 with a pharmaceutically acceptable diluent to a set volume.

54. The composition of claim 53, wherein the pharmaceutically acceptable diluent is a sugar solution, optionally wherein the pharmaceutically acceptable diluent is a dextrose solution, optionally wherein the

pharmaceutically acceptable diluent is 5% Dextrose Injection, USP.

55. The composition of any one of claims 53-54, wherein the set volume is 250 mL.

56. The composition of any of claims 53-55, wherein the concentration of I-66 is about 0.05-10 mg/mL, or about 0.05-6 mg/mL, or about 0.1-5 mg/mL.

57. The composition of any one of claims 53-56, wherein the concentration of the solubilizing agent, amino acid, or arginine is about 0.05-10 mg/mL, or about 0.05-6 mg/mL, or about 0.01-5 mg/mL.

58. The composition of any one of claims 53-57, wherein the concentration of the surfactant or PEG is about 0.01%-3% w/v, or about 0.05%-2% w/v, or about 0.01%-3% v/v, or about 0.05%-2% v/v.

59. The composition of any one of claims 53-58, wherein the concentration of the buffer agent or buffer is about 0.1-10 mM, or about 0.2-5 mM.

60. The composition of any one of claims 53-59, wherein the buffer agent is sodium phosphate monobasic, or wherein the buffer agent is sodium phosphate monobasic, or wherein the buffer is a phosphate buffer, or wherein the buffer is a Tris buffer.

61. The composition of any one of claims 53-60, wherein the pharmaceutically acceptable diluent is 5% Dextrose Injection, USP and the concentration of dextrose is about 0.02-0.05 mg/mL.

62. A composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;

a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally a pH adjustment agent;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and
water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about

0.1-10 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% w/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% w/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% w/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and

water; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, comprising:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
a buffer; and
dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;
a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;
a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

a buffer agent, wherein the concentration of the buffering agent is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally a pH adjustment agent;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;
optionally sodium hydroxide;
optionally hydrochloric acid;
dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and
water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;
arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.01-100 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.1-10 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.01-0.05 mg/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;

arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;

sodium phosphate monobasic, wherein the concentration of sodium phosphate monobasic is about 0.2-5 mM;

optionally sodium hydroxide;

optionally hydrochloric acid;

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; and

water; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

an amino acid, wherein the concentration of the amino acid is about 0.1-6 mg/mL;

a PEG or a surfactant, wherein the concentration of the PEG or surfactant is about 0.01%-3% v/v;

a buffer; and

dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.1-6 mg/mL;

arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;

PEG-400, wherein the concentration of PEG-400 is about 0.01%-3% v/v;
 a buffer; and
 dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or
 a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
 arginine, wherein the concentration of arginine is about 0.1-6 mg/mL;
 PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
 a buffer; and
 dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or
 a composition, consisting of:

I-66, wherein the concentration of I-66 is about 0.2-5 mg/mL;
 arginine, wherein the concentration of arginine is about 0.2-5 mg/mL;
 PEG-400, wherein the concentration of PEG-400 is about 0.05%-2% v/v;
 a buffer; and
 dextrose, wherein the concentration of the dextrose is about 0.02-0.05 g/mL; or

63. The composition of claim 62, wherein:
 the concentration of I-66 is about 0.2-5 mg/mL;
 the concentration of arginine is about 0.2-5 mg/mL;
 the concentration of PEG-400 is about 0.06-1.7% w/v; and
 the concentration of dextrose is about 0.02-0.05 mg/mL; or

wherein:

the concentration of I-66 is about 0.2-5 mg/mL;
 the concentration of arginine is about 0.2-5 mg/mL;
 the concentration of PEG-400 is about 0.06-1.7% v/v; and
 the concentration of dextrose is about 0.02-0.05 mg/mL.

64. The composition of any one of claims 62-63, wherein the buffer is a phosphate buffer or a Tris buffer, and/or wherein the concentration of the buffer is about 0.01-10 mM or about 0.02-5 mM.

65. The composition of any of the claims 62-64, wherein the pH of the composition is about 7.0-9.0, or wherein the pH of the composition is about 8.0.

66. The composition of any one of claims 62-65, wherein I-66 is Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and wherein:

¹³C NMR of I-66 in MeOD at 25°C, comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5,

51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58, and I-66 has a purity of about or at least about 90%; and/or

¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise, or ¹H NMR of I-66 in MeOD at 25°C comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

67. The composition of any one of claims 62-66, wherein identity of I-66 is confirmed by comparison with a reference standard, wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOH⁶-Aib⁷-Ala⁸-Phc⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ^1H NMR in MeOD at 25°C, its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ^1H NMR comprises the following peaks (chemical shift (δ , ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise.

68. The composition of claim 67, wherein the comparison is or comprises utilizing HPLC, optionally wherein the HPLC condition is selected from those in Example 1, as described in Table E-7 or as described in Table E-10.

69. The composition of any one of claims 67-68, wherein the comparison is or comprises utilizing NMR, optionally wherein the NMR is ^1H NMR, ^{13}C NMR, and/or two- or multi-dimensional NMR.

70. The composition of any one of the preceding claims, wherein I-66 has a purity of about 80%, 85%, 95%, or more.

71. A method, comprising diluting a composition of I-66 with a pharmaceutically acceptable diluent, optionally wherein the method comprises:

calculating the volume of a composition of I-66 for administration of an intended dose;

removing from an infusion bag/container containing a pharmaceutically acceptable diluent a volume of the diluent equivalent to the calculated volume; and

addition of the calculated volume of the composition of I-66 to the infusion bag/container; and optionally wherein:

the concentration of the I-66 in the composition is about 1-25 mg/mL, about 5-25 mg/mL, or about 10 mg/mL;

the composition of I-66 is a composition of any one of claims 1-54; and/or

the pharmaceutically acceptable diluent is 5% Dextrose Injection, USP;

optionally wherein the method prepares an I-66 for administration, optionally intravenous administration; and optionally wherein the method prepares a composition of any one of claims 55-72.

72. A method, comprising comparing I-66 in a preparation with a reference standard, optionally wherein the reference standard is an I-66 preparation wherein I-66 has the structure of Ac-PL3¹-Asp²-Npg³-B5⁴-Asp⁵-3COOHF⁶-Aib⁷-Ala⁸-Phe⁹-Lys¹⁰-PyrS2¹¹-3Thi¹²-BztA¹³-Glu¹⁴-Ala¹⁵-NH₂, wherein there are olefin staples between PL3¹ and B5⁴ and between B5⁴ and PyrS2¹¹ and a lactam staple between Lys¹⁰ and Glu¹⁴, and:

when characterized by ¹³C NMR in MeOD at 25°C, its ¹³C NMR comprises the following peaks (chemical shift (δ, ppm)): 17, 17.76, 23.2, 23.2, 23.29, 23.32, 23.87, 24.46, 27.46, 27.47, 28.98, 29.61, 29.99, 30, 30.18, 30.63, 31.36, 31.76, 31.81, 32.6, 33.09, 33.8, 33.84, 35.69, 35.8, 35.96, 36.18, 36.46, 38.1, 38.18, 39.25, 45.37, 45.96, 49.5, 51.2, 53.94, 54.54, 55.33, 55.56, 55.69, 56.48, 56.93, 57.28, 57.7, 59.94, 60.7, 60.94, 62.78, 65.89, 67.18, 70.51, 122.71, 123.42, 123.87, 125.19, 125.2, 125.37, 126.96, 126.99, 128.08, 129.15, 129.2, 129.37, 129.54, 129.83, 129.83, 130.39, 130.39, 131.26, 131.47, 131.96, 132.12, 132.57, 135.08, 137.36, 138.06, 138.13, 139.58, 141.98, 156.51, 169.51, 172.38, 172.48, 173.13, 174.1, 174.39, 174.47, 175.44, 175.55, 175.55, 175.6, 175.7, 175.8, 175.8, 175.8, 176.97, 177, 177.56, 178.08, and 178.58; and

when characterized by ¹H NMR in MeOD at 25°C, its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, and 2.23; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H),

2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), and 2.23 (3H), wherein each peak represents 1H unless indicated otherwise; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93, 2.38, 2.92, 3.45, 1.18, 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37, 2.38, 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2, 2.42, 2.22, 3.08, 3.40, 1.55, 1.07, 1.42, 1.59, 1.33, 2.23, 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69; or its ¹H NMR comprises the following peaks (chemical shift (δ, ppm)): 3.96, 6.34, 6.84, 6.36, 4.38, 4.31, 3.82, 4.21, 7.21, 7.24, 7.07, 7.24, 7.21, 4.03, 4.25, 7.97, 7.94, 7.42, 7.5, 4.4, 5.29, 5.31, 4.27, 4.31, 5.56, 5.36, 7.89, 7.38, 7.41, 7.89, 7, 4.3, 2.00, 2.10, 1.60, 1.65, 3.73, 4.15, 3.15, 4.22, 2.29, 2.41, 3.53, 3.25, 3.06, 2.95, 1.93 (2H), 2.38 (2H), 2.92, 3.45, 1.18 (2H), 1.36, 1.55, 1.84, 1.53, 3.27, 3.31, 3.27, 3.44, 2.74, 2.96, 1.92, 2.07, 1.37 (2H), 2.38 (2H), 1.80, 1.88, 2.71, 2.81, 3.20, 2.05, 1.97, 2.17, 0.84, 1.42, 1.57, 2.69, 3.76, 3.92, 2.2 (2H), 2.42, 2.22, 3.08, 3.40, 1.55 (3H), 1.07 (3H), 1.42 (3H), 1.59 (3H), 1.33 (3H), 2.23 (3H), 8.32, 7.73, 8.17, 8.19, 8.25, 8.44, 8.89, 8.1, 8.42, 7.45, 7.39, 8.15, 8.86, and 7.69, wherein each peak represents 1H unless indicated otherwise;

optionally wherein the comparison is or comprises utilizing HPLC, optionally wherein the HPLC condition is selected from those in Example 1, as described in Table E-7 or as described in Table E-10; and

optionally wherein the comparison is or comprises utilizing NMR, optionally wherein the NMR is ¹H NMR, ¹³C NMR, and/or two- or multi-dimensional NMR;

optionally wherein the method confirms identity of I-66 in the preparation;

optionally wherein:

the method is utilized to control quality the I-66 preparation; and/or

the method is utilized to release the I-66 preparation;

optionally wherein:

the I-66 preparation is a preparation of I-66 drug substance; or

the I-66 preparation is a preparation of I-66 drug product; and

optionally wherein:

I-66 in the preparation is in or comprises a pharmaceutically acceptable salt form, or I-66 in the preparation is in or comprises a sodium salt form, or I-66 in the reference is in or comprises a pharmaceutically acceptable salt form, or I-66 in the reference is in or comprises a sodium salt form.

73. A method for modulating beta-catenin interaction with a partner in a system, comprising administering or delivering to the system I-66, optionally wherein the partner is TCF4; or

a method for modulating a TCF-beta-catenin interaction in a system, comprising contacting beta-catenin with I-66; or

a method for modulating a TCF-beta-catenin interaction in a system, comprising administering or delivering to the system I-66; or

a method for inhibiting beta-catenin dependent cell proliferation, comprising administering or delivering to the system I-66; or

a method for decreasing beta-catenin polypeptide level in a system, comprising administering or delivering to the system I-66; or

a method for decreasing c-Myc polypeptide and/or transcript level in a system, comprising administering or delivering to the system I-66, optionally wherein c-Myc polypeptide level is reduced and/or c-Myc mRNA level is reduced, and/or optionally wherein the reduction is observed 1, 2, 3, 4, 5, 6, 7, 8, 9, or 10 or more days post a dose; or

a method for decreasing Axin2 polypeptide and/or transcript level in a system, comprising administering or delivering to the system I-66; optionally wherein Axin2 polypeptide level is reduced and/or Axin2 mRNA level is reduced; or

a method for modulating WNT/beta-catenin pathway in a system, comprising administering or delivering to the system I-66, wherein expression of a nucleic acid is modulated; or

a method, comprising administering or delivering to the system I-66, wherein level of a transcript of a nucleic acid and/or a product thereof is modulated; or

a the method the I-66 preparation method, comprising administering or delivering to the system I-66, wherein expression of a nucleic acid is modulated.

74. The method of claim 73, wherein:

a nucleic acid is or comprises a gene.

a system is an in vitro system;

a system is an in vivo system;

a system is or comprises a sample;

a system is or comprises a cell, tissue or organ;

a system is or comprises cancer cells;

a system is or comprises a tumor; and/or

a system is a subject.

75. A method for treating cancer, comprising administering or delivering to a subject suffering therefrom an effective amount of I-66.

76. The method of claim 75, wherein the cancer is a solid tumor.

77. The method of claim 75, wherein the cancer is colorectal cancer, gastric cancer, lung cancer, non-small cell lung cancer, metastatic non-small cell lung cancer, non-small cell lung cancer stage IIIB, non-small cell carcinoma, non-small cell carcinoma of lung, TNM Stage 4, or gastroesophageal-junction cancer, or wherein the cancer is advanced or metastatic MSS colorectal cancer, advanced or metastatic non-small cell lung cancer, advanced or metastatic gastric cancer, or advanced or metastatic gastroesophageal junction cancer, or wherein the cancer is advanced or metastatic non-small cell lung cancer with documented WPAM in APC, advanced or metastatic non-small cell lung cancer with documented WPAM in beta-catenin,

advanced or metastatic gastric cancer with documented WPAM in APC, advanced or metastatic gastric cancer with documented WPAM in beta-catenin, advanced or metastatic gastroesophageal junction cancer with documented WPAM in APC, or advanced or metastatic gastroesophageal junction cancer with documented WPAM in beta-catenin, optionally wherein the cancer comprises a mutation selected from *APC* loss of function mutations, *CTNNB1* gain of function mutations, *RNF43* LOF mutations, *RSPO2* and *RSPO3* fusions, or a mutation selected from mutations in *AMER1*, *AXIN1*, *AXIN2*, *BCL9*, *CSNK1A1*, *GSK3B*, *LRP5*, *LRP6*, *LGR5*, *TCF7L2*, and *WIF1*.

78. The method of any one of claims 75-77, wherein:

- the tumor is non-MSI-H;
- the tumor is non-dMMR;
- the cancer is a locally advanced cancer;
- the cancer is a metastatic cancer; and/or
- the cancer is microsatellite stable.

79. The method of any one of claims 75-78, wherein the cancer comprises a Wnt pathway activating mutation (WPAM), the cancer comprises an APC mutation and/or a beta-catenin mutation, and/or the cancer is refractory.

80. The method of any one of claims 73-79, comprising:

- administering or delivering I-66 weekly;
- administering or delivering I-66 about weekly for at least about three or four doses;
- administering or delivering I-66 about weekly in continuous cycles of 21 or 28 days;
- administering or delivering I-66 in a composition having a volume of about 250 mL;
- administering or delivering I-66 in a composition of any one of claims 1-72; and/or
- administering or delivering I-66 intravenously.

81. The method of any one of claims 73-80, wherein:

- a dose of about 18 mg/m² I-66 is administered;
- a dose of about 36 mg/m² I-66 is administered;
- a dose of about 72 mg/m² I-66 is administered;
- a dose of about 144 mg/m² I-66 is administered;
- a dose of about 240 mg/m² I-66 is administered;
- a dose of about 360 mg/m² I-66 is administered;
- a dose of about 480 mg/m² I-66 is administered; and/or
- a dose of about 600 mg/m² I-66 is administered.

82. The method of any one of the preceding claims, comprising administering or deliver to a subject a second therapeutic agent or second therapy.

83. A composition or method of any one of Embodiments 1-398.

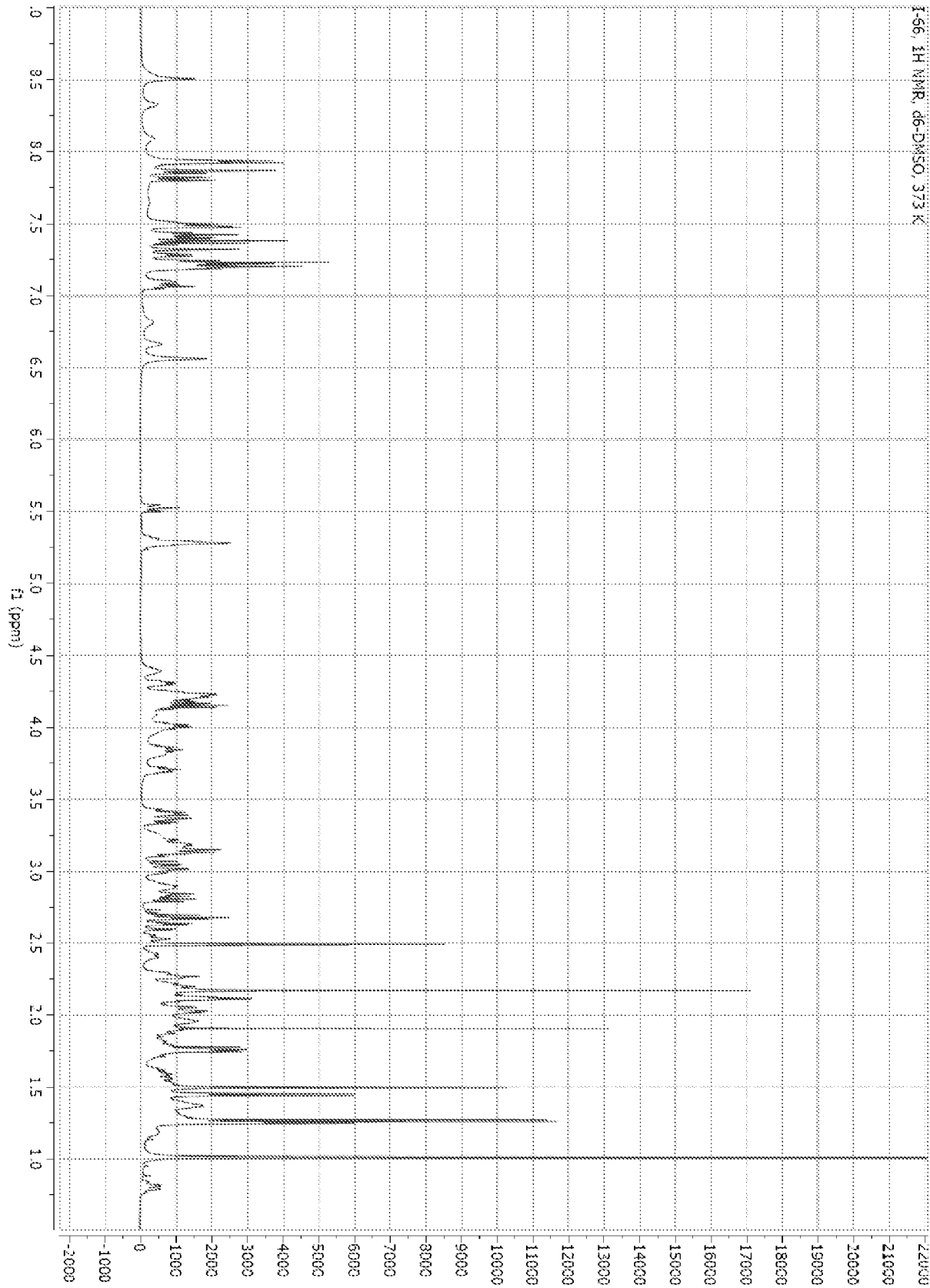


FIG. 1

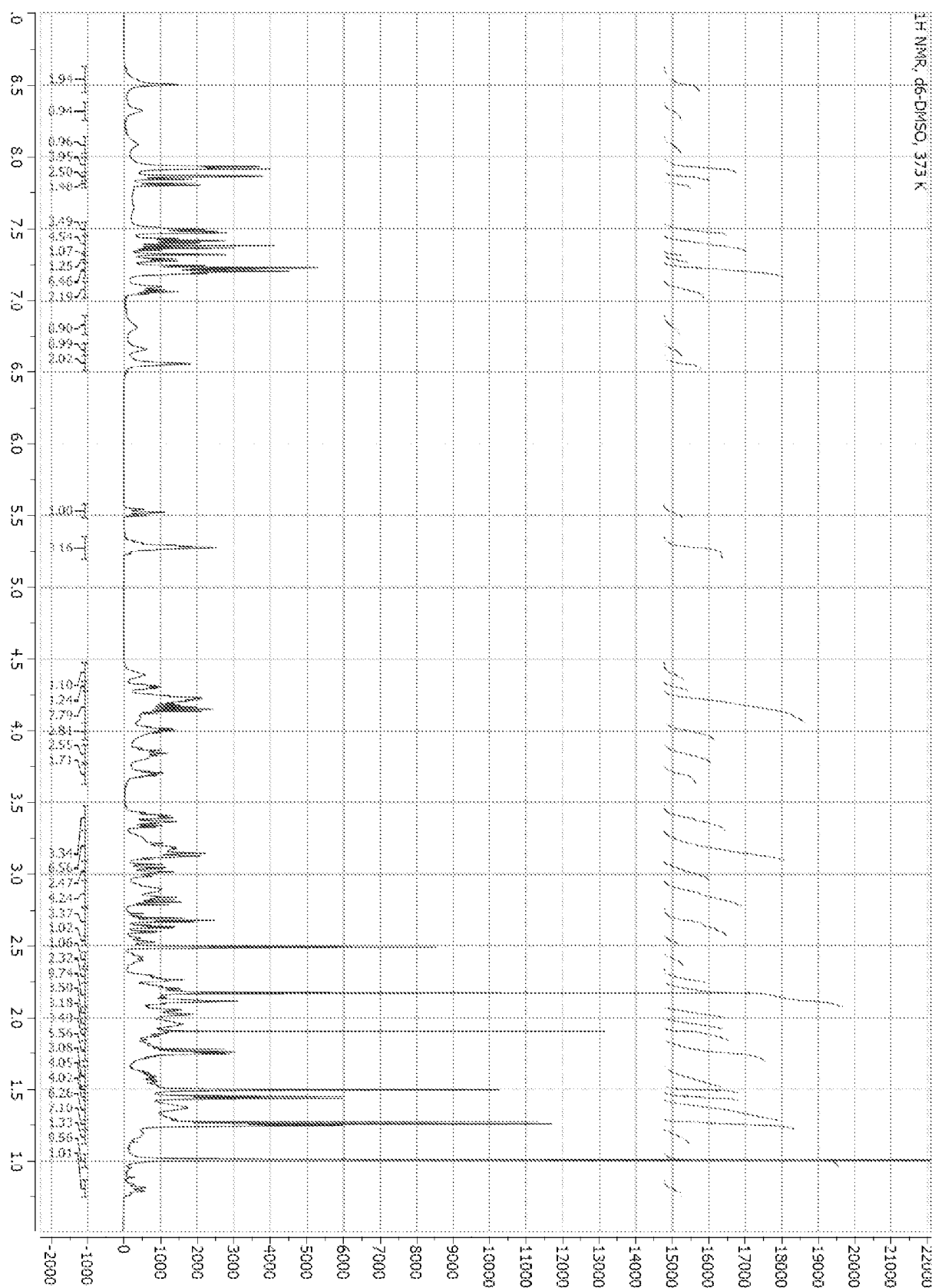
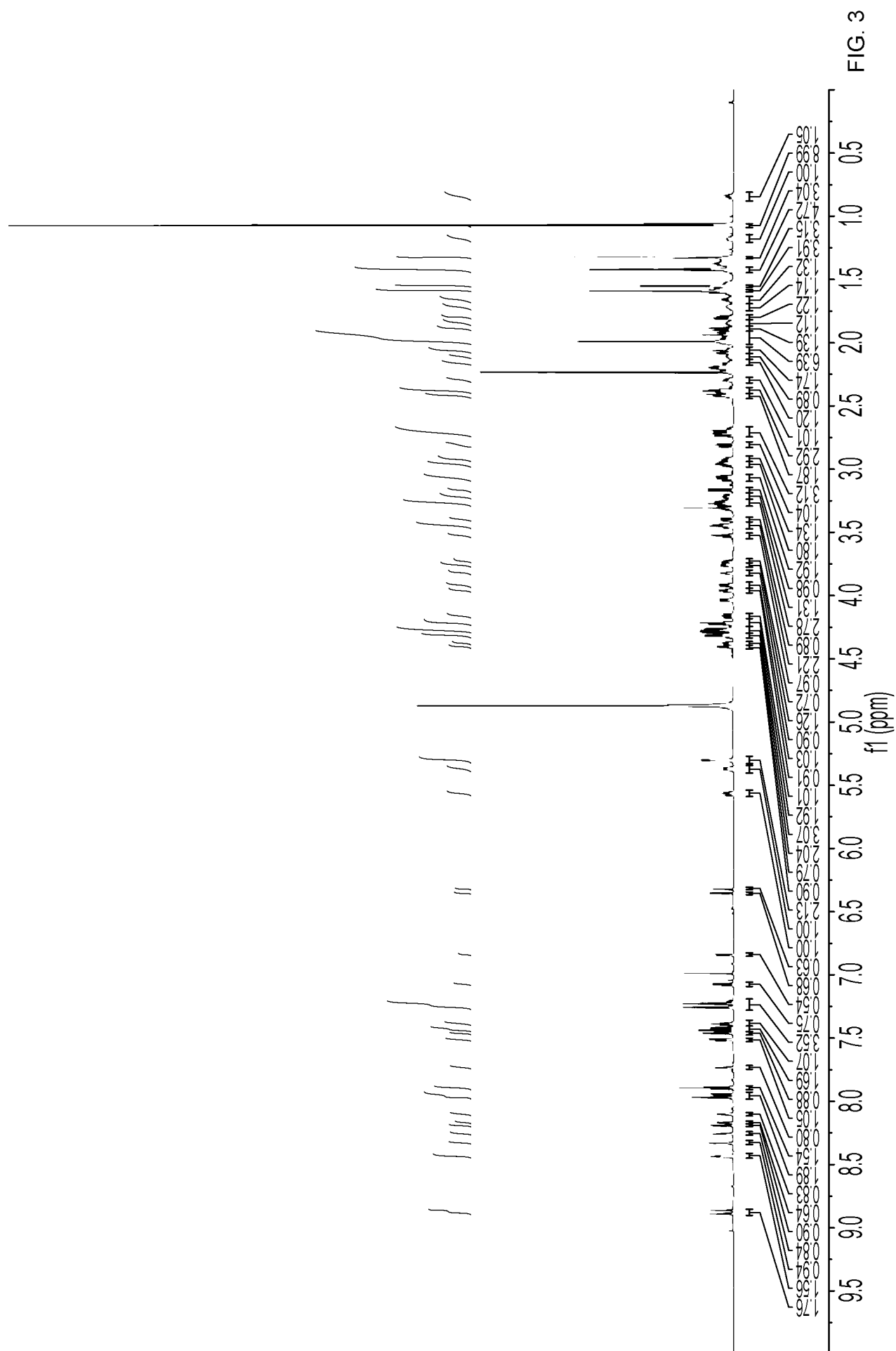


FIG. 2



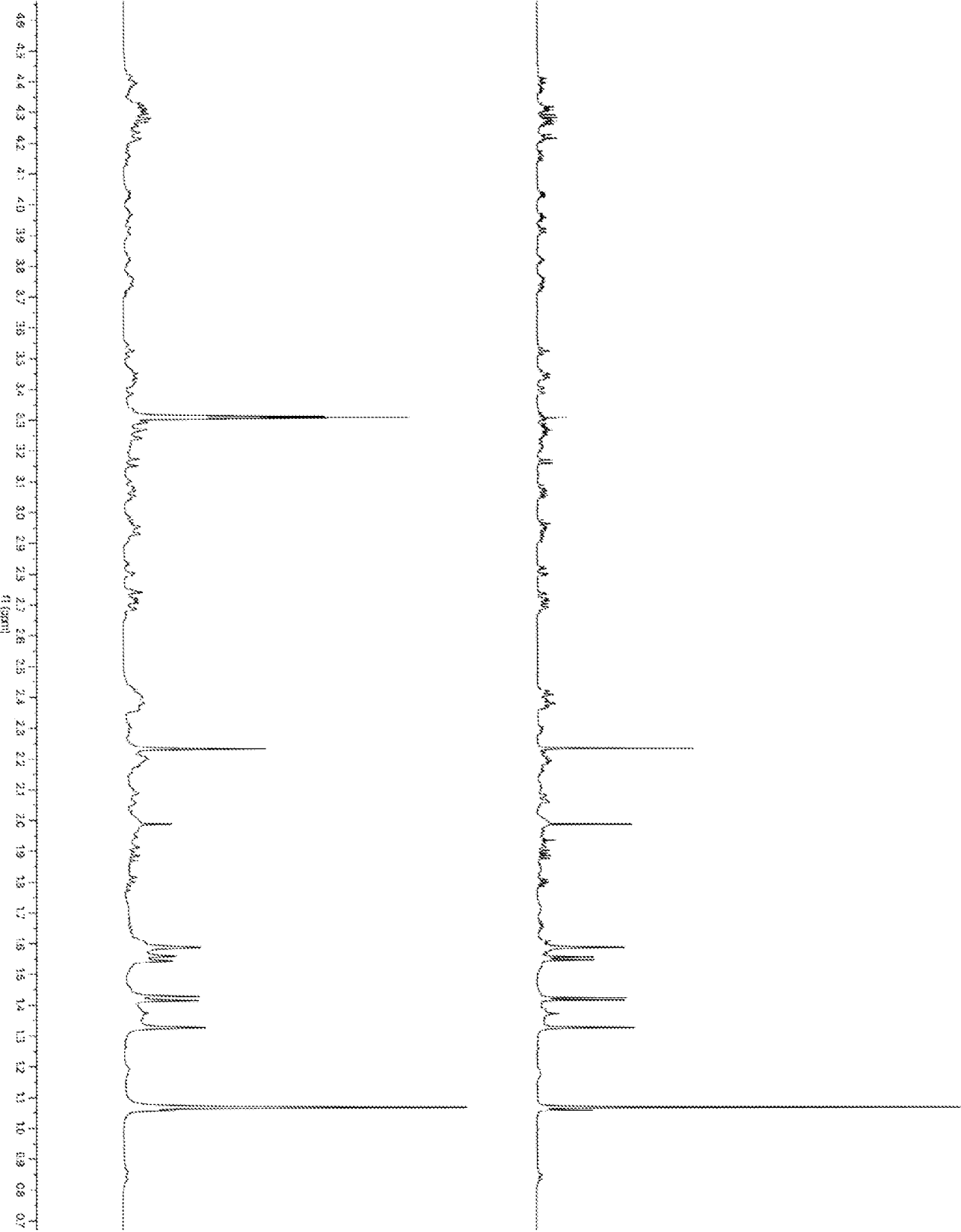


FIG. 4

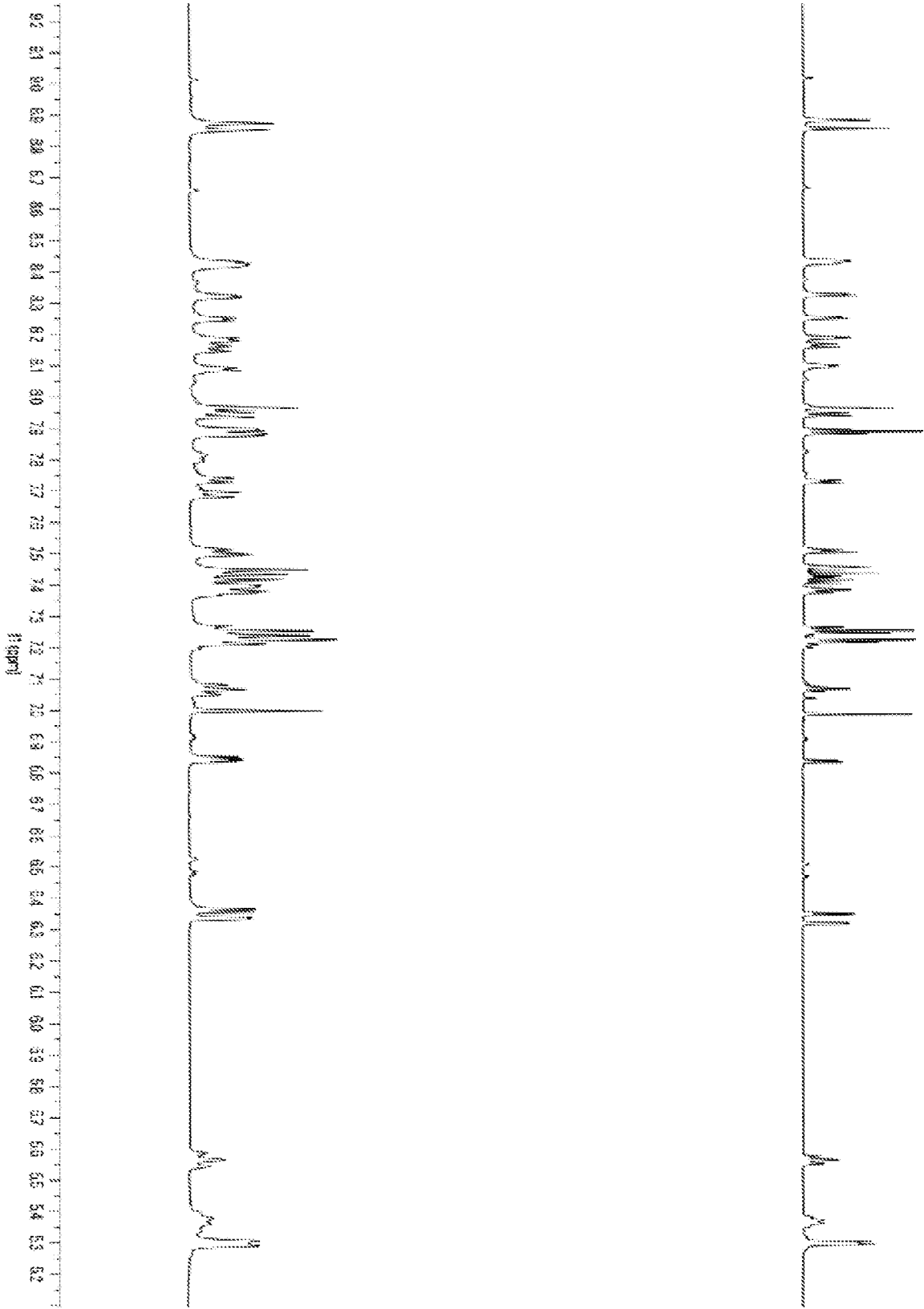


FIG. 5

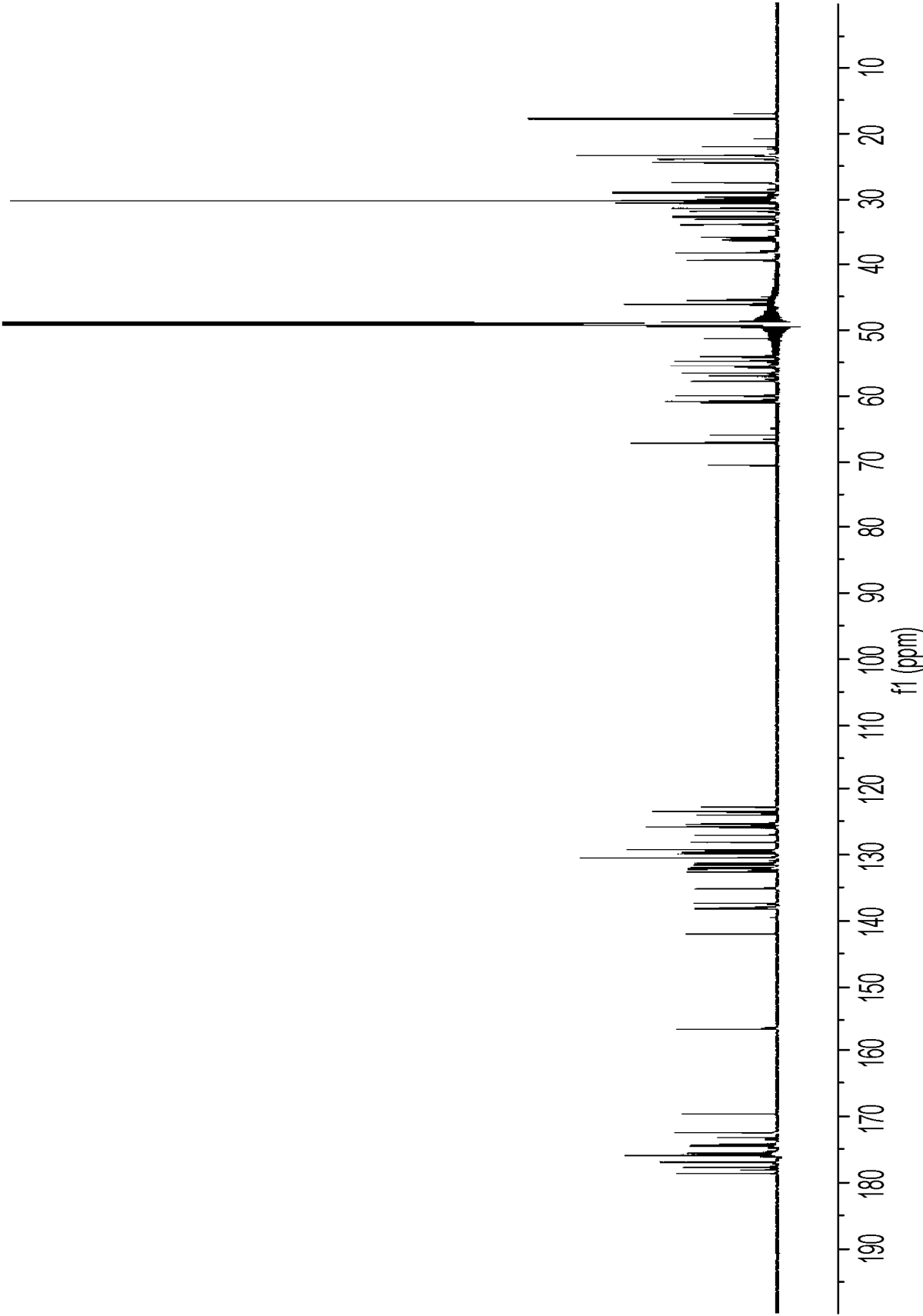


FIG. 6

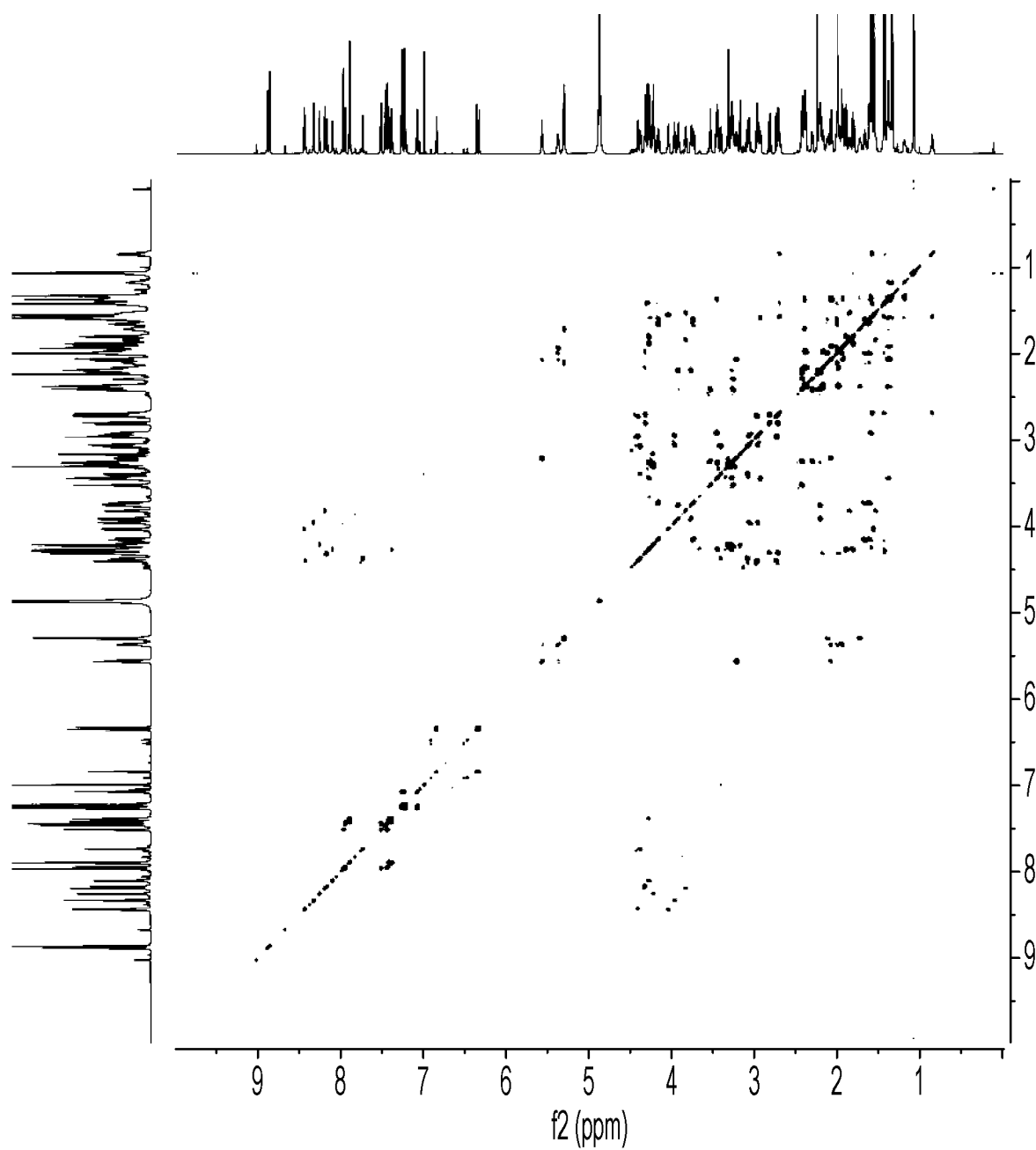


FIG. 7

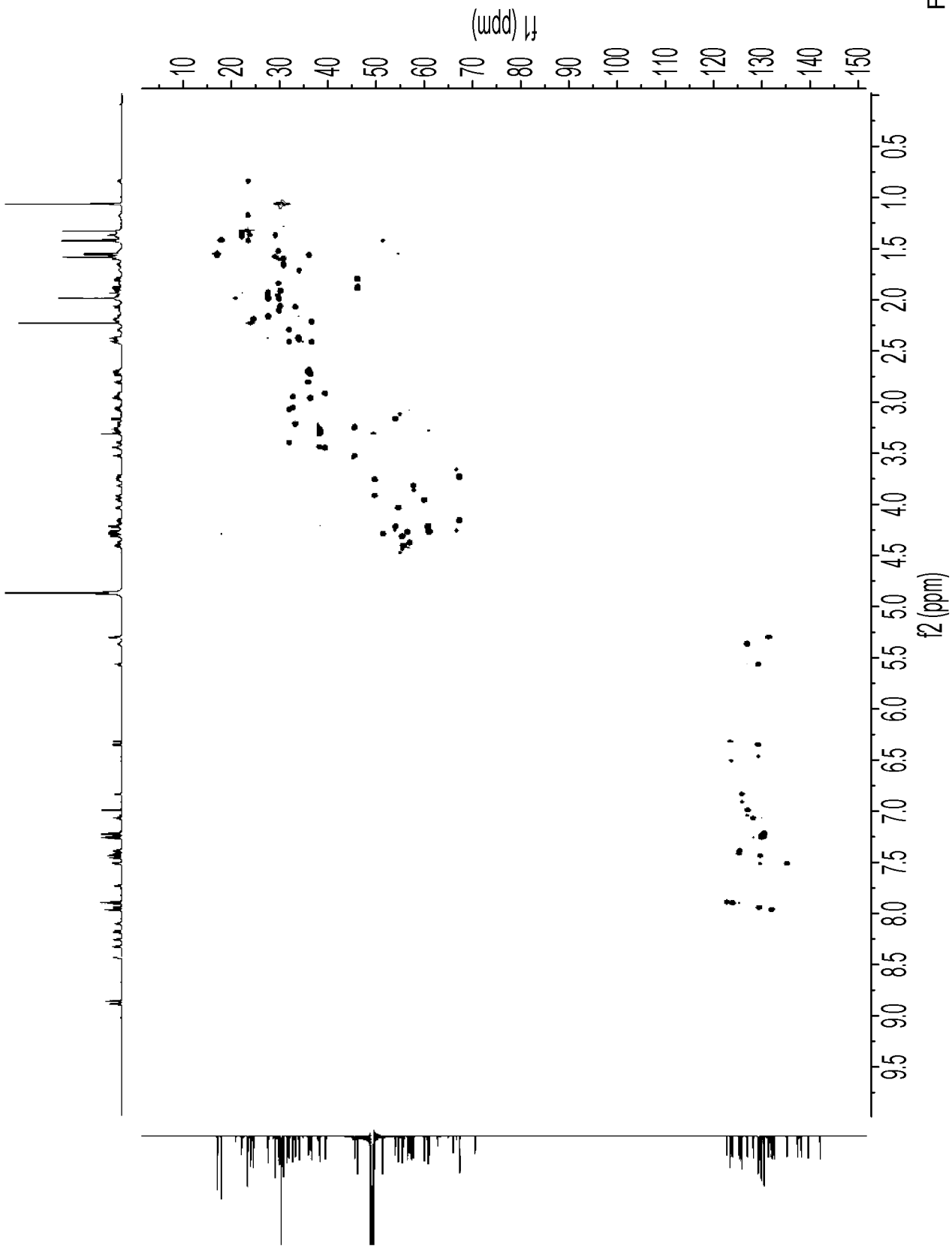


FIG. 8

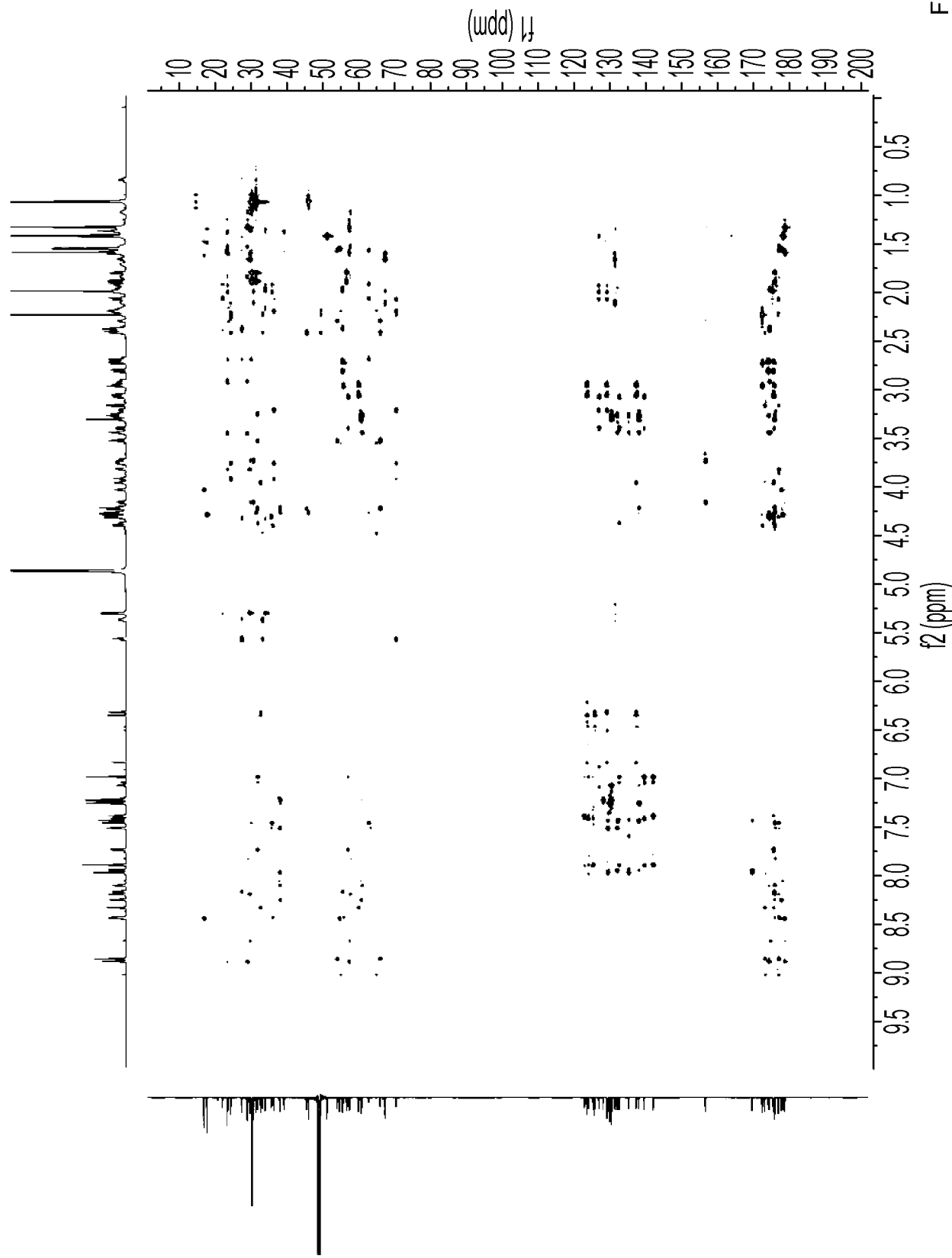


FIG. 9

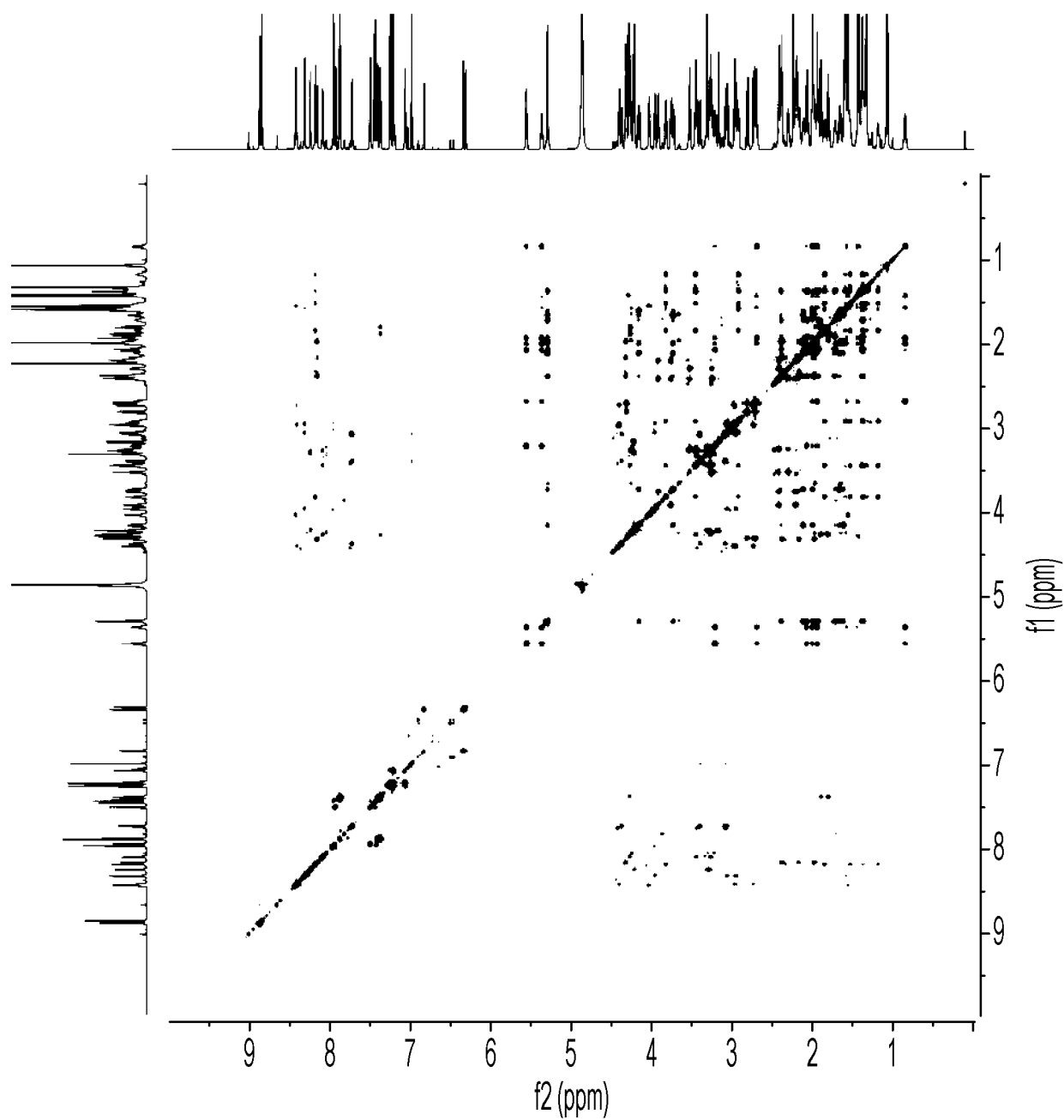


FIG. 10

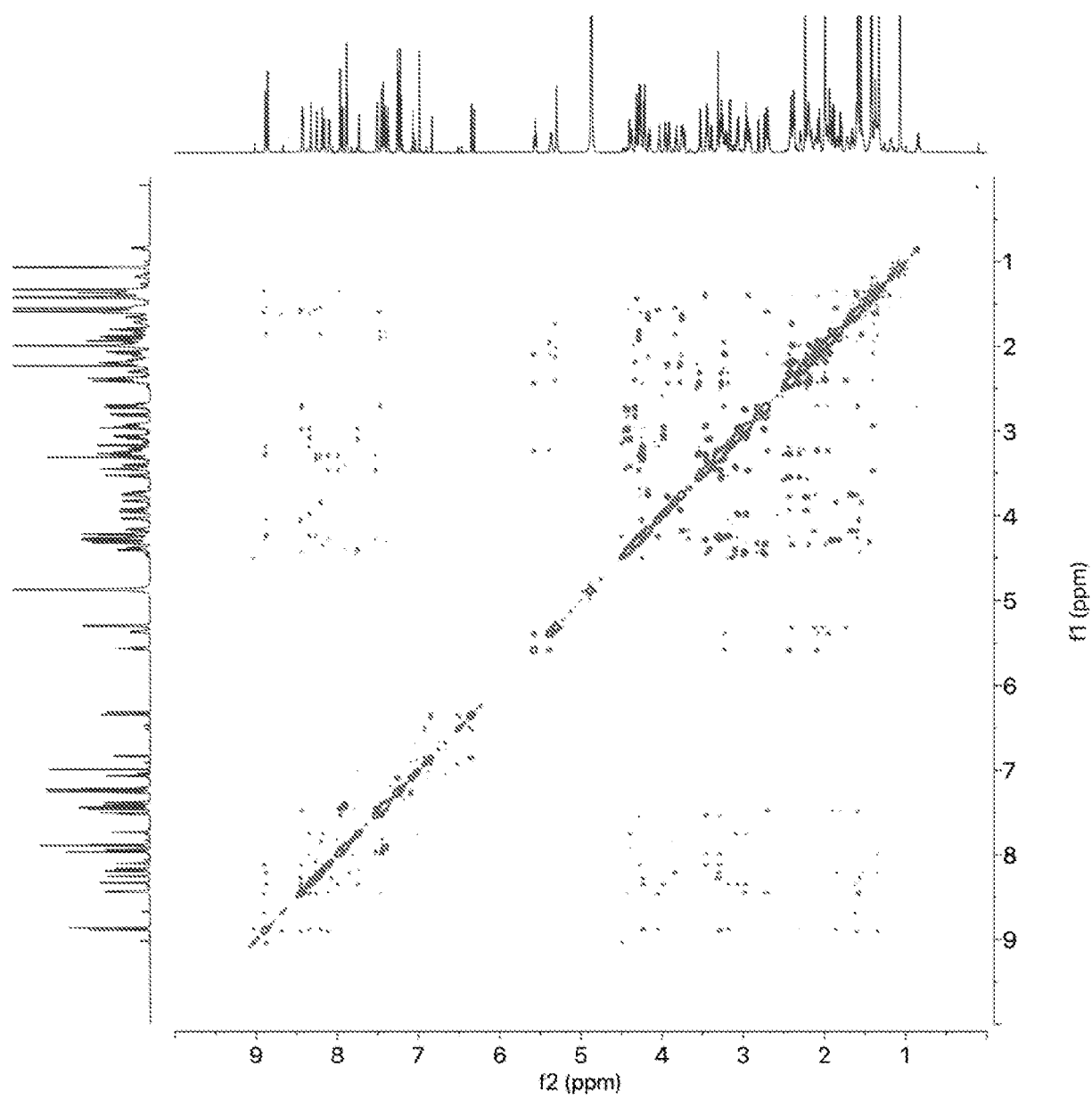


FIG. 11

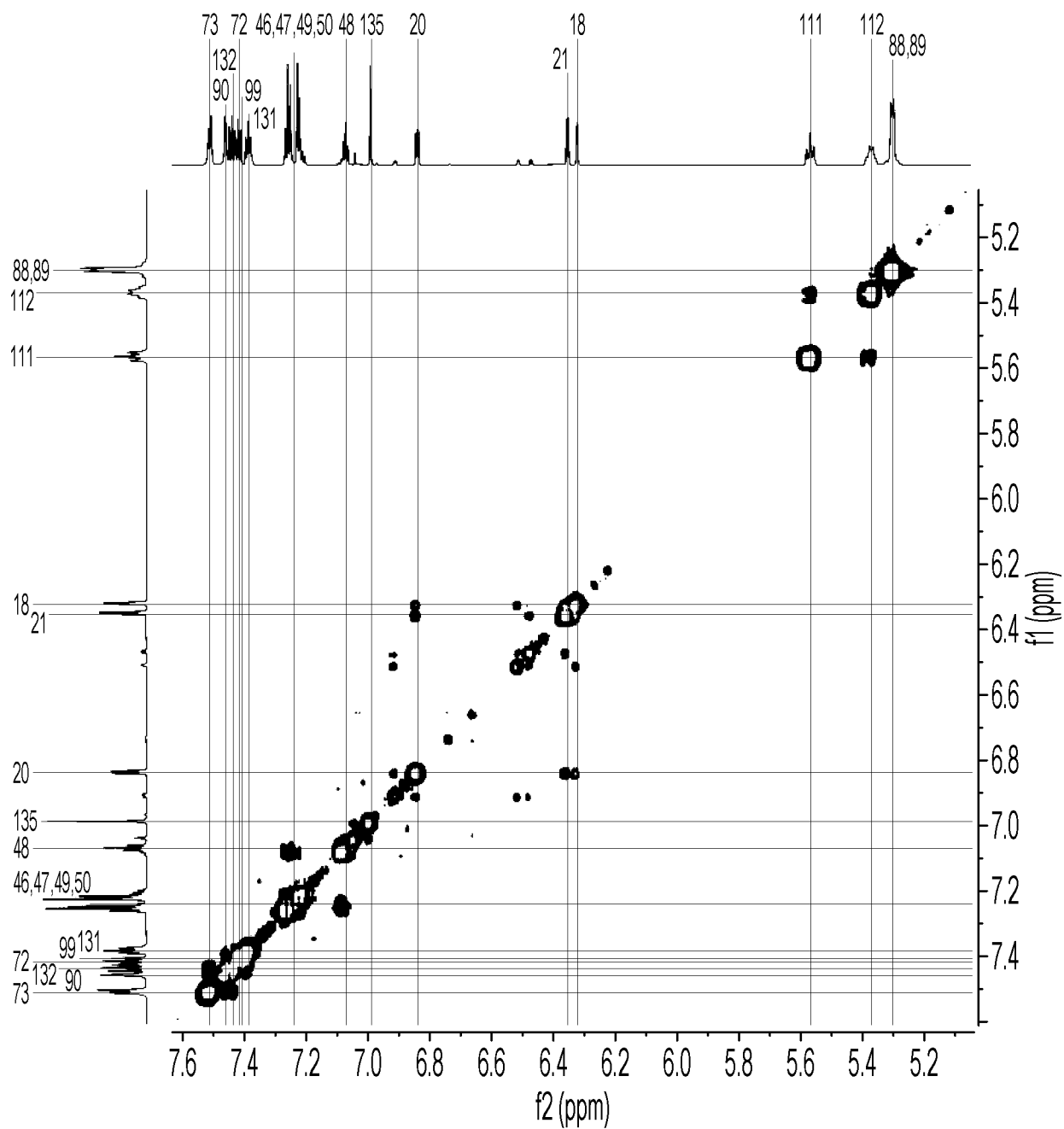


FIG. 12

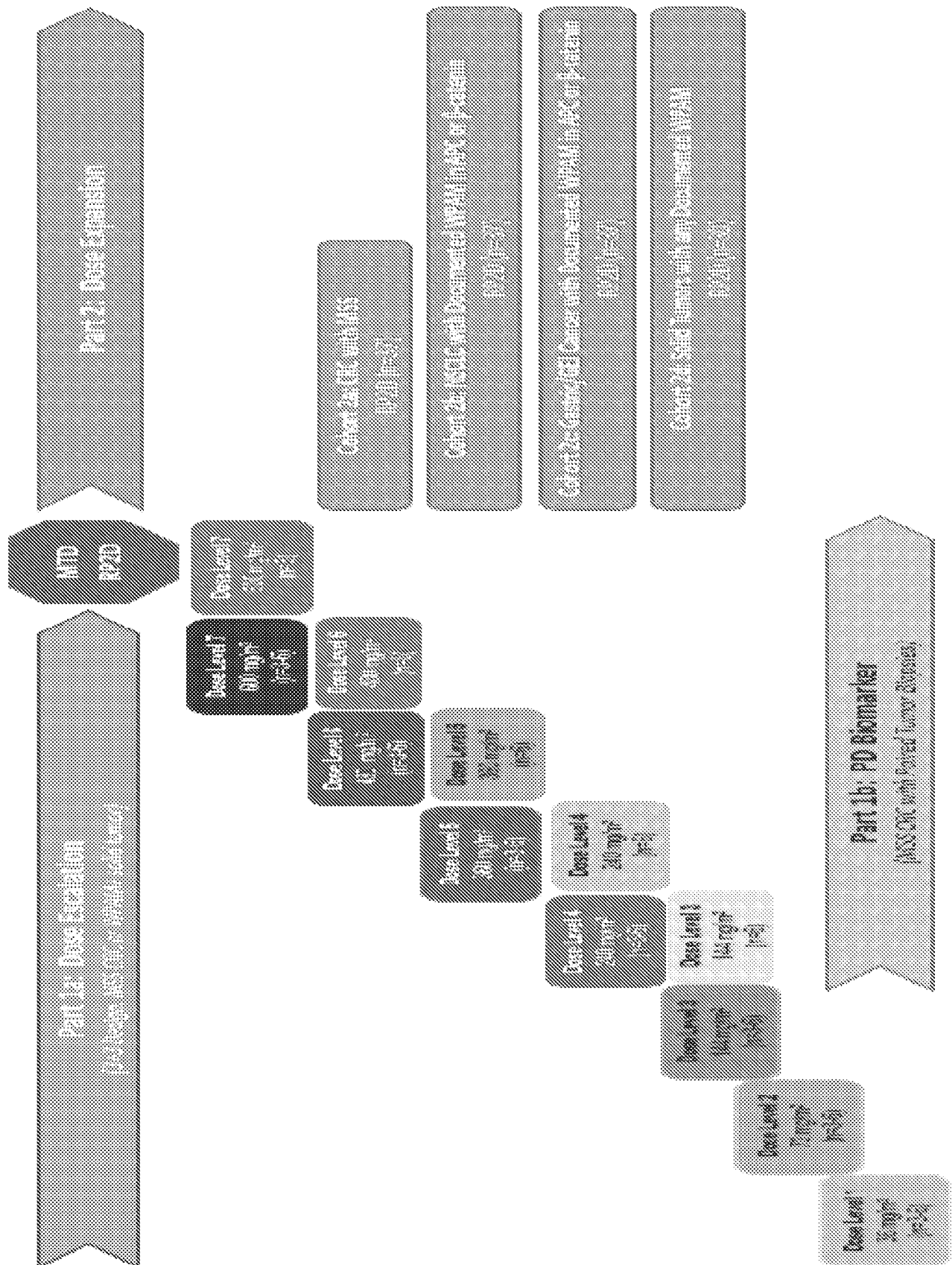


FIG. 13

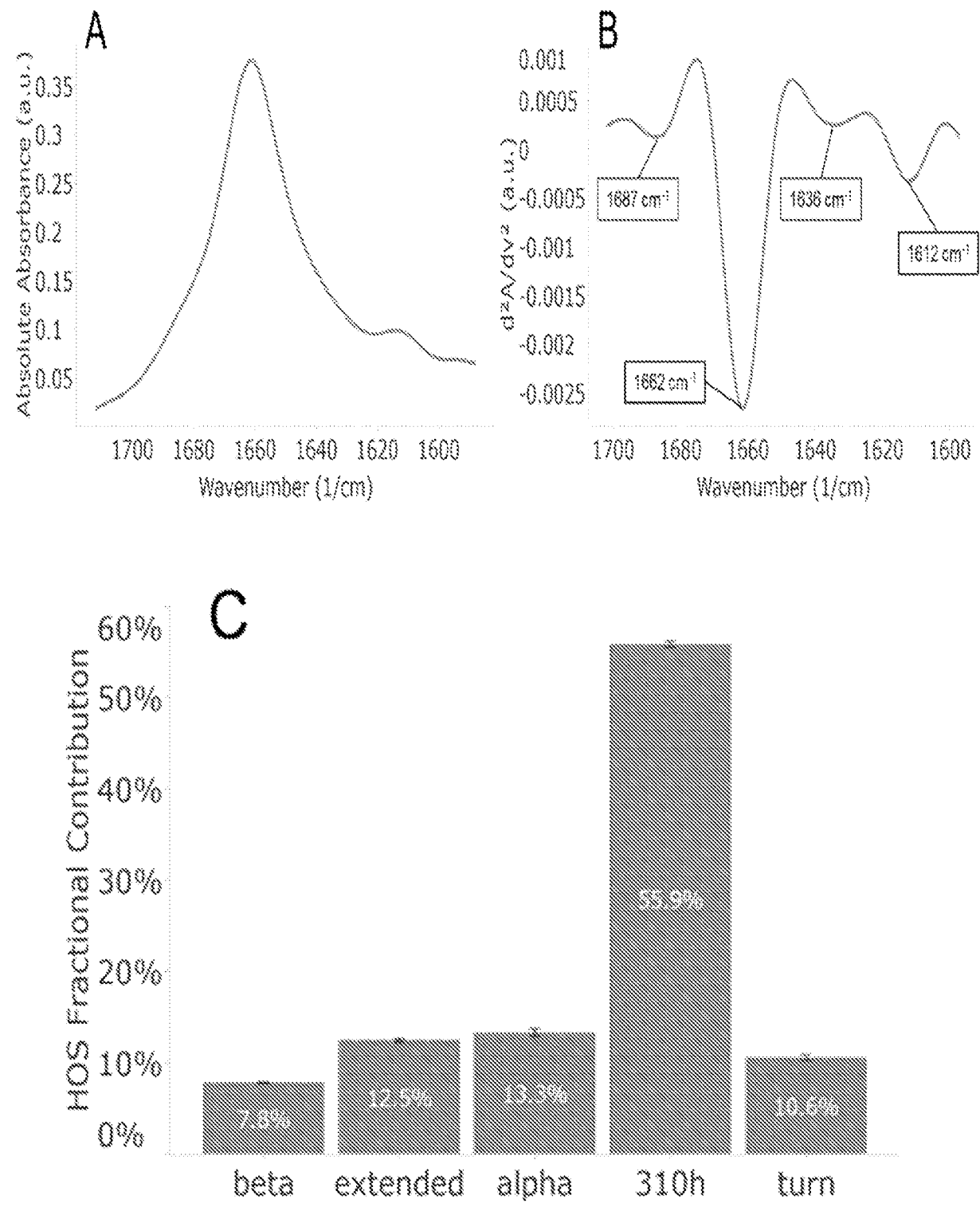


FIG. 14