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- FIG. 1D

- [Continued on next page]

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ZA, ZM, ZW.

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INTRACELLULAR TARGETING OF OLIGONUCLEOTIDES

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims the benefit of priority to the filing date of U.S. Provisional Application Serial No. 63/520,274, filed August 17, 2023; U.S. Provisional Application Serial No. 63/520,271, filed August 17, 2023; U.S. Provisional Application Serial No. 63/557,137, filed February 23, 2024; U.S. Provisional Application Serial No. 63/561,529, filed March 5, 2024; U.S. Provisional Application Serial No. 63/561,552, filed March 5, 2024; U.S. Provisional Application Serial No. 63/640,478, filed April 30, 2024; and U.S. Provisional Application Serial No. 63/665,630, filed June 28, 2024, the disclosure of each of which is specifically incorporated by reference herein in its entirety.

BACKGROUND

[0002] Biologic macromolecules, such as proteins and nucleic acids hold enormous potential as therapeutic agents, especially against targets that are challenging for conventional small molecule drug modalities. However, a major problem in bringing such therapies to the clinic is their limited ability to gain access to the intracellular compartment when administered systemically. Carrier systems, such as polymers, cationic liposomes or chemical modifications, have been used facilitate intracellular delivery of macromolecular therapeutics. Still, intracellular delivery efficiency by these approaches is often low and improved delivery systems to increase efficacy of intracellular delivery have remained elusive.

[0003] The present disclosure addresses this and other issues.

SUMMARY

[0004] One strategy to subvert the membrane barrier and deliver biologic therapeutics, such as nucleic acids, into cells is to attach them to “cell-penetrating peptides” (also referred to herein as CPPs or EEVs (endosomal escape vehicles), for example, as disclosed in International Application Publication No. WO 2022/213118, filed March 31, 2022, entitled CYCLIC CELL PENETRATING PEPTIDES, and WO 2022/241408, filed May 9, 2022, entitled COMPOSITIONS AND METHODS FOR MODULATING TISSUE DISTRIBUTION OF

INTRACELLULAR THERAPEUTICS, the disclosures of which are hereby incorporated by reference in their entireties.

[0005] While not wishing to be bound by theory, it is believed that the linkers can influence efficacy and tolerability of the resultant construct, for example to increase efficacy.

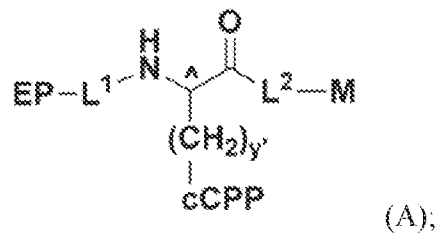
5 [0006] Therapeutics can be conjugated to CPPs or EEVs using a variety of chemistries, including, for example, click chemistry or amide chemistry. Although both methods are effective at conjugating the therapeutic to the EEV, the different chemistries result in different linkages that may impart different properties to the resulting molecule. EEV generated using click chemistry for cargo conjugation include a hydrophobic component at the point of conjugation
10 that is absent when amide chemistry is used. While not wishing to be bound by theory, molecules that include the hydrophobic component at the point of conjugation appear to have improved efficacy as compared to similar molecules in which the therapeutic is conjugated to the EEV using amide chemistry. However, there are problems associated with click chemistry that are not experienced with amide chemistry, for example, formation of regioisomers. While amide
15 chemistry eases the synthetic process, it results in the loss of the hydrophobic component at the point of conjugation. EEVs incorporating a hydrophobic component are provided herein that demonstrate enhanced efficacy as compared to a similar EEVs in which the hydrophobic component is absent. In embodiments, the EEVs incorporating the hydrophobic component do not show any decrease in tolerability as compared to the similar EEV in which the hydrophobic
20 component is absent.

[0007] While not wishing to be bound by theory, it is also believed that the linker length can influence efficacy and tolerability. In some instances, as linker length decreases, efficacy increases and tolerability decreases; and as linker length increases, efficacy decreases and tolerability increases. EEVs incorporating different linker lengths are provided herein.

25 Additionally, it is believed that including one or more amino acid residues in the linker can influence efficacy and tolerability. EEVs incorporating one or more amino acid residues are provided herein. Furthermore, it is believed that the chirality of the amino acids in a cell-penetrating peptide (CPP) or endosomal escape vehicle (EEV) can impact cytosolic delivery efficiency, as well as toxicity.

30 [0008] An EEV can comprise a cyclic cell penetrating peptide (cCPP), an exocyclic peptide (EP) and trivalent linker comprising the structure of Formula (A):

Formula (A):



wherein:

EP is a linear exocyclic peptide;

5 cCPP is a cyclic cell penetrating peptide;

L^1 and L^2 , are, independently, a linker arm;

^ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5; and

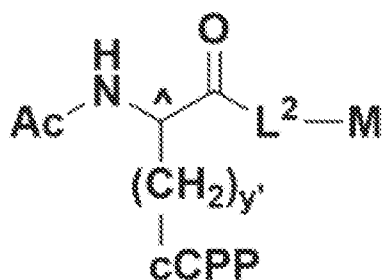
M comprises a reactive handle.

10 [0009] In embodiments, L^1 and L^2 , are each, independently, absent, or comprise hydrocarbon component (e.g., $\text{NRH}-(\text{CH}_2)_n-\text{COOH}$), a polyethylene glycol (PEG) component, a hydrophobic component (X), an amino acid component comprising one or more amino acid residues (AA), or any combination thereof.

[0010] In embodiments, L^1 for Formula (A) comprises (AA) a' or (PEG) x' , any combination thereof, and L^2 comprises one or more amino acid components (AA) b' , (AA) b'' , one or more PEG components (PEG) z' , (PEG) z'' , one or more hydrophobic components (e.g., $\text{X}^{o'}$, $\text{X}^{o''}$, etc.) or any combination thereof, wherein (AA) a' , (AA) b' , and (AA) b'' are independently an amino acid component comprising at least one amino acid residue.

15 [0011] In embodiments, the EEV can comprise a cyclic cell penetrating peptide (cCPP) and a linker. In embodiments, the EEV does not include a linear exocyclic peptide (EP). In embodiments, the EEV has the structure shown in Formula (AA):

Formula (AA):



(AA);

wherein:

Ac is acetyl;

5 cCPP is a cyclic cell penetrating peptide;

L¹ and L², are, independently, a linker arm;

^ indicates L- or D-stereochemistry;

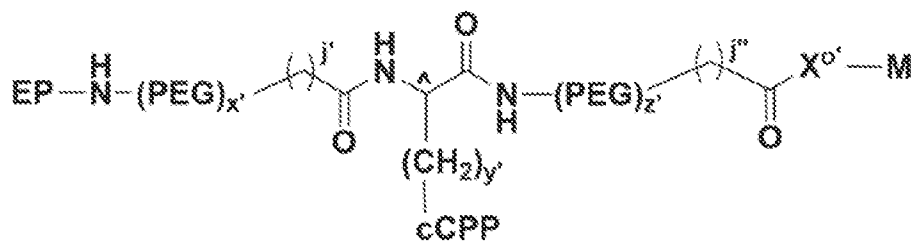
y' is an integer from 1 to 5; and

M comprises a reactive handle.

- 10 [0012] In embodiments, the linker arm L² of Formula (AA), comprises one or more amino acid components (e.g., (AA)b', (AA)b'', etc.), one or more PEG components (e.g., (PEG)z', (PEG)z'', etc.), one or more hydrophobic components (e.g., X^{o'}, X^{o''}, etc.), or any combination thereof, wherein (AA)b' and (AA)b'' are independently an amino acid component comprising at least one amino acid residue, b' and b'' are independently an integer from 0-12, and z' and z'' are
- 15 independently an integer from 0-12.

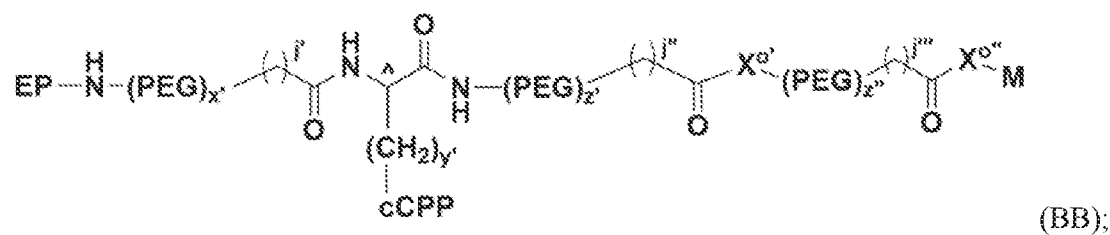
[0013] In embodiments, the EEV of Formula (A) comprises a structure selected from:

Formula (B):

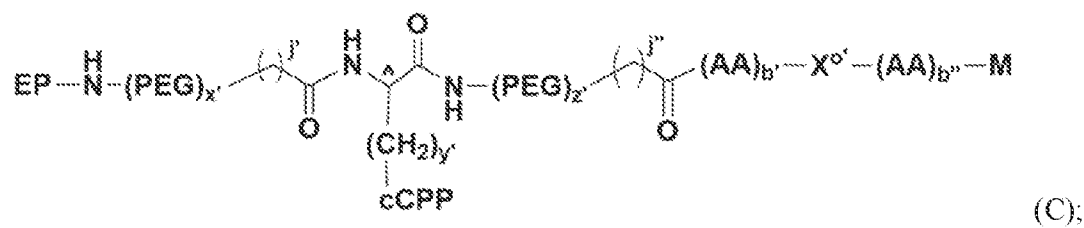


(B);

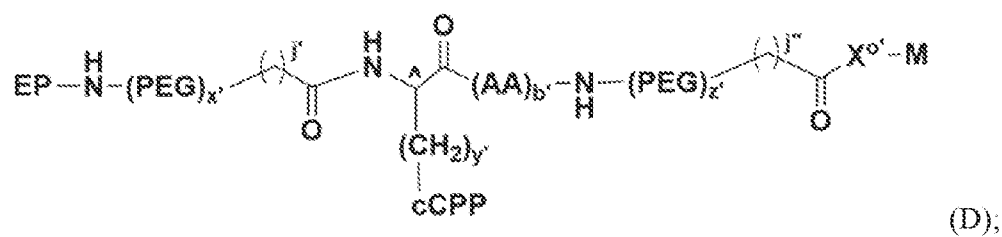
Formula (BB):



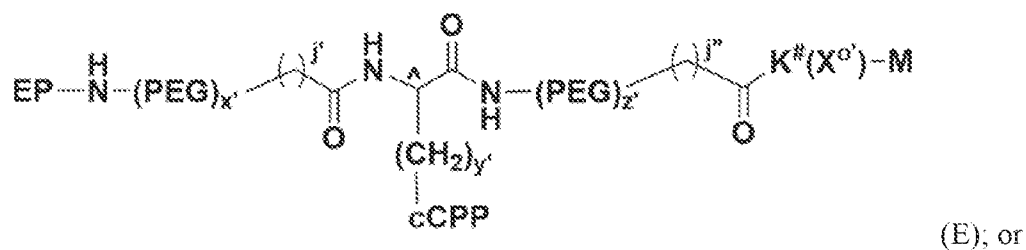
Formula (C):



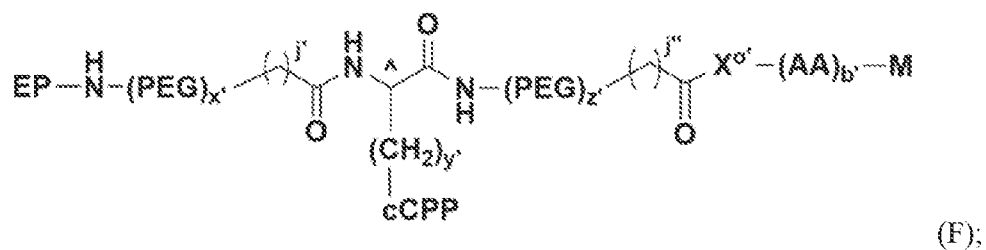
5 Formula (D):



Formula (E):



Formula (F):



10

wherein:

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

5 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ and $X^{o''}$ are each, independently, a hydrophobic component;

10 $K^\#$ is D-lysine or L-lysine residue;

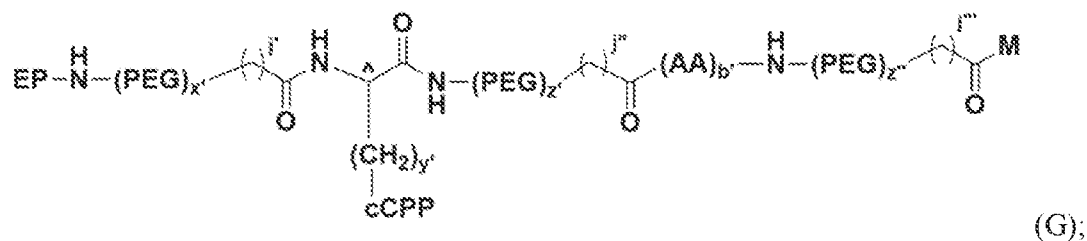
$(AA)^{b'}$ and $(AA)^{b''}$ are each an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

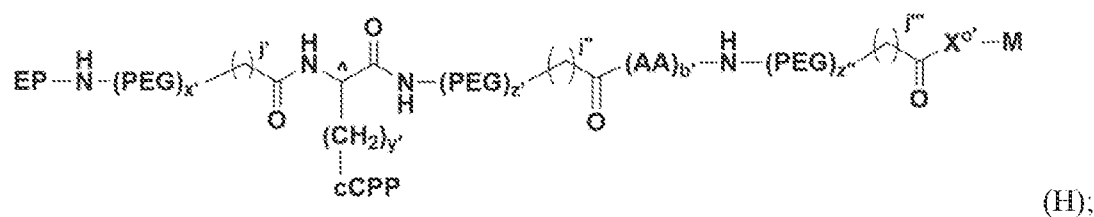
b'' is an integer from 0 to 12.

15 [0014] In embodiments, the EEV of Formula (A) comprises a structure selected from:

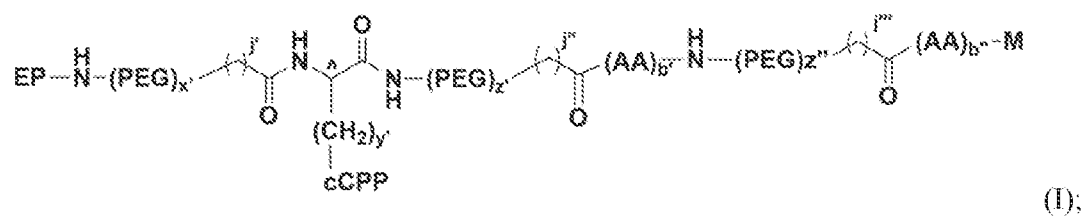
Formula (G):



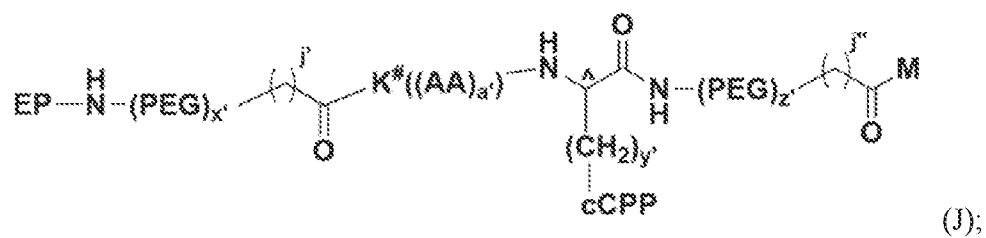
Formula (H):



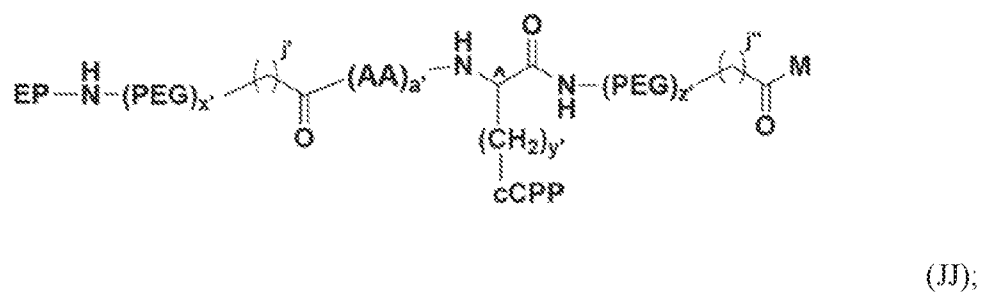
Formula (I):



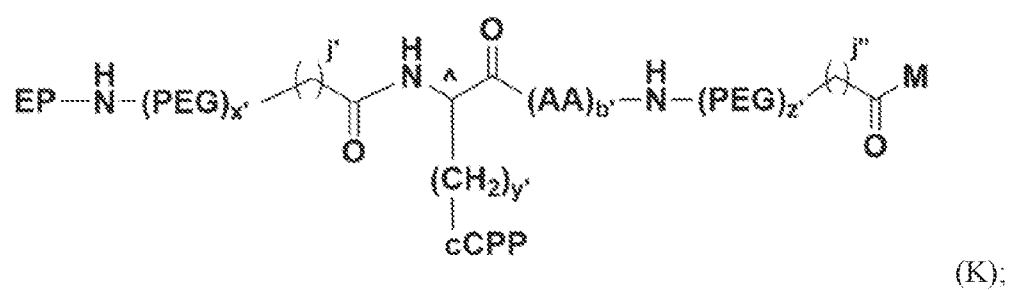
Formula (J):



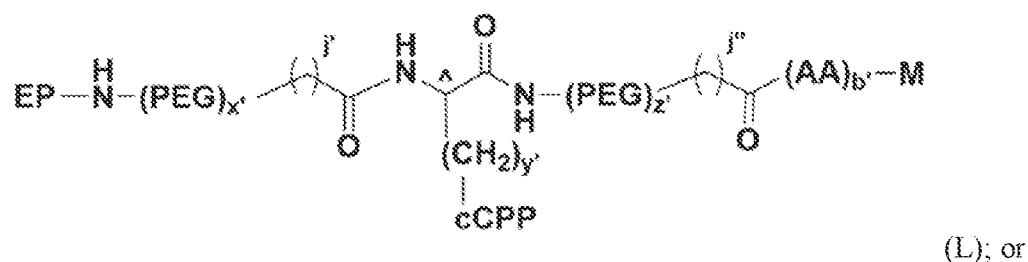
5 Formula (JJ):



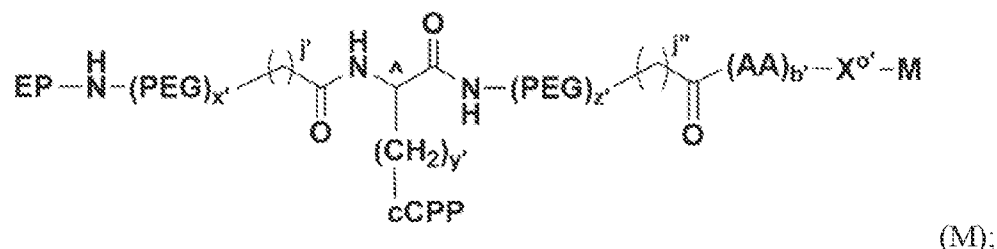
Formula (K):



Formula (L):



Formula (M):



5 wherein:

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

10 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ comprises a hydrophobic component;

15 $K^{\#}$ is D-lysine or L-lysine residue;

$(AA)a'$, $(AA)b'$, and $(AA)b''$ are each an amino acid (AA) component comprising at least one amino acid residue;

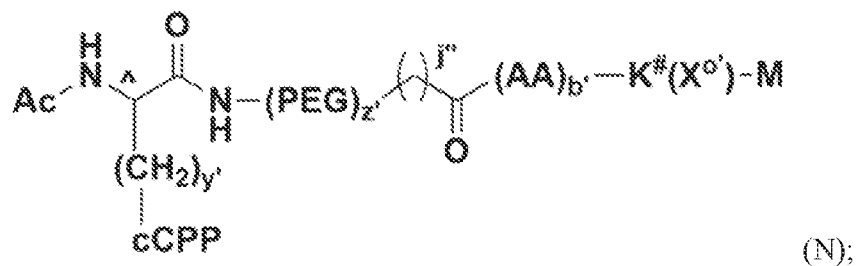
a' is an integer from 0 to 12;

b' is an integer from 0 to 12; and

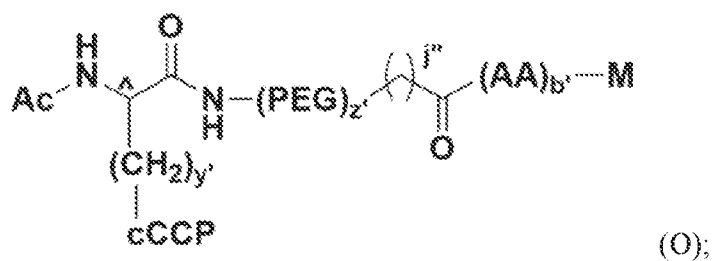
20 b'' is an integer from 0 to 12.

[0015] In embodiments, the EEV of Formula (AA) comprises a structure selected from:

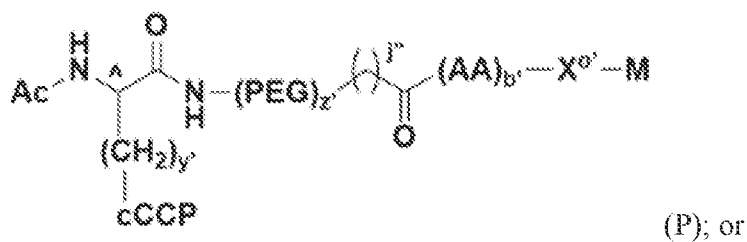
Formula (N):



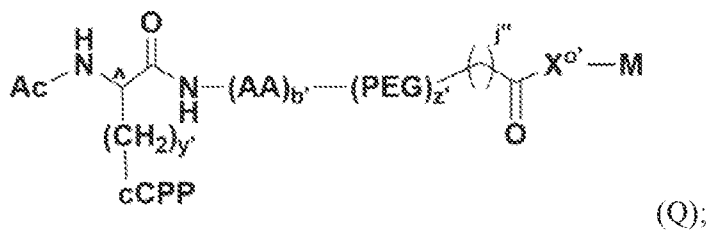
Formula (O):



5 Formula (P):



Formula (Q):



wherein:

- 10 Ac is acetyl;
 cCPP is a cyclic cell penetrating peptide;
 z' is an integer from 0 to 12;
 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
 X^{o'} is a hydrophobic component;

$K^{\#}$ is D-lysine or L-lysine residue;

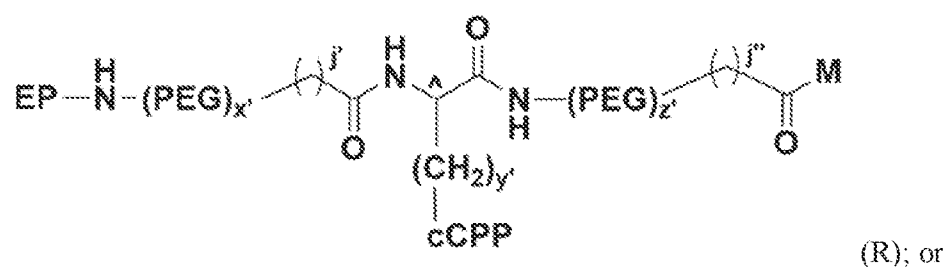
(AA) b' is an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

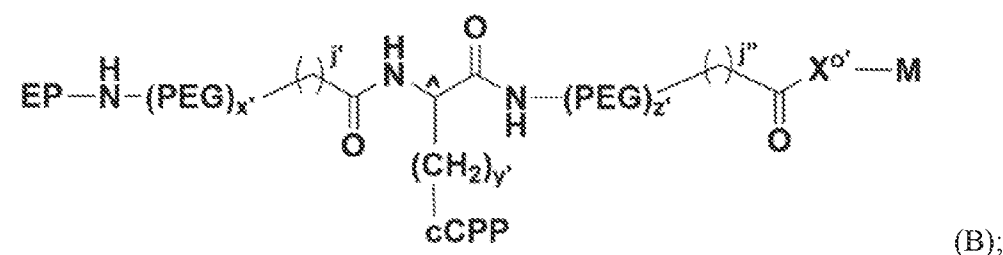
[0016] In embodiments, the EEV comprises a structure selected from Formula (R) and Formula

5 (B):

Formula (R):



Formula (B):



10 wherein

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

15 ^ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$X^{o'}$ comprises a hydrophobic component; and

20 M comprises a reactive handle.

[0017] An EEV can comprise a cyclic cell penetrating peptide (cCPP), an exocyclic peptide (EP) and linker, wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to

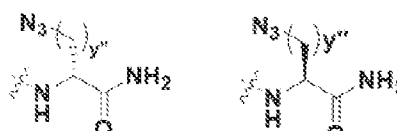
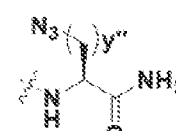
the linker. In embodiments, cCPP comprises at least 2 D-amino acids with a hydrophobic side chain and at least 2 D-arginine amino acids. In embodiments, cCPP comprises at least 2 D-phenylalanine. In embodiments, L² comprises a hydrophobic component (X^{o'}). In embodiments, X^{o'} is Nal, dNal, or BiP. In embodiments, AAsc is a side chain of glutamine. In embodiments, AAsc is a side chain of L-glutamine. In embodiments, AAsc is a side chain of D-glutamine.

[0018] In embodiments, the EEV comprises a structure selected from Formula (B), (BB), (C), (D), (E), or (F), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0019] In embodiments, the EEV comprises a structure selected from Formula (G), (H), (I), (J), (JJ), (K), (L), or (M), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

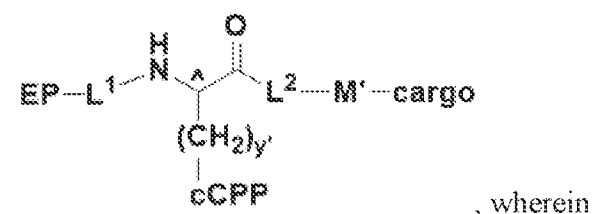
[0020] In embodiments, the EEV comprises a structure selected from Formula (N), (O), (P), or (Q), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0021] In embodiments, the EEV comprises a structure selected from Formula (R) or (B), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0022] In embodiments, M comprises -OH, , or , wherein y'' is an integer from 1-4.

[0023] In embodiments, the EEV can be conjugated to a cargo to form an EEV-cargo conjugate comprising the structure of Formula (A-1):

Formula (A-1):



EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

L^1 and L^2 , are, independently, a linker arm;

^ indicates L- or D-stereochemistry;

5 y' is an integer from 1 to 5;

M' comprises a bonding group; and

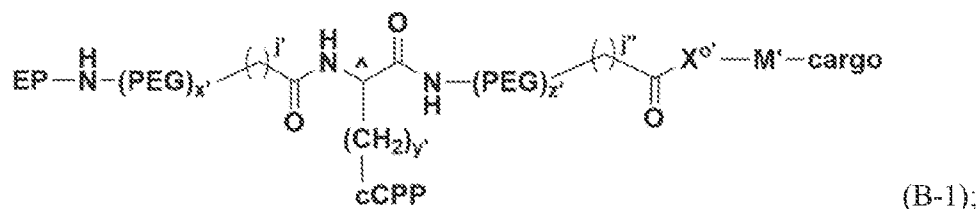
cargo is a peptide, an oligonucleotide, a small molecule, or any combinations thereof.

[0024] In embodiments, L^1 and L^2 , are each, independently, absent, or comprise one or more hydrocarbon components (e.g., $\text{NRH}-(\text{CH}_2)_n-\text{COOH}$), one or more polyethylene glycol components, one or more hydrophobic components, one or more amino acid components, or any combination thereof.

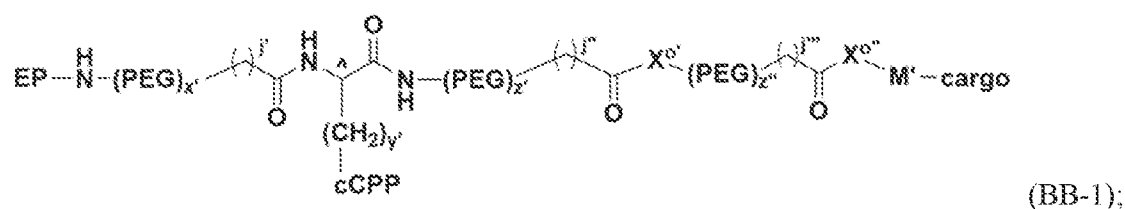
[0025] In embodiments, L^1 comprises $(\text{AA})a'$ or $(\text{PEG})x'$, or any combination thereof, and L^2 comprises $(\text{AA})b'$, $(\text{AA})b''$, $(\text{PEG})z'$, $(\text{PEG})z''$, one or more hydrophobic components (e.g., $X^{o'}$, $X^{o''}$, etc.), or any combination thereof.

15 [0026] In embodiments, the compound of Formula (A-1) comprises a structure selected from:

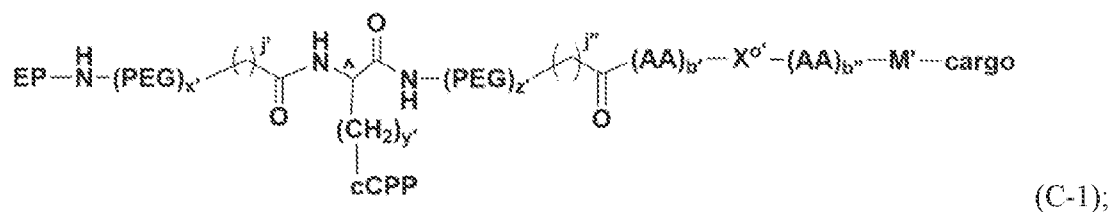
Formula (B-1):



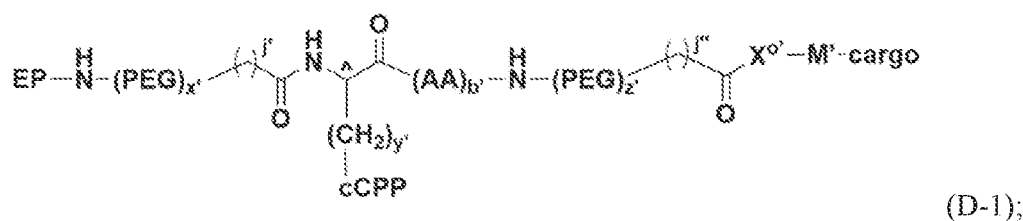
Formula (BB-1):



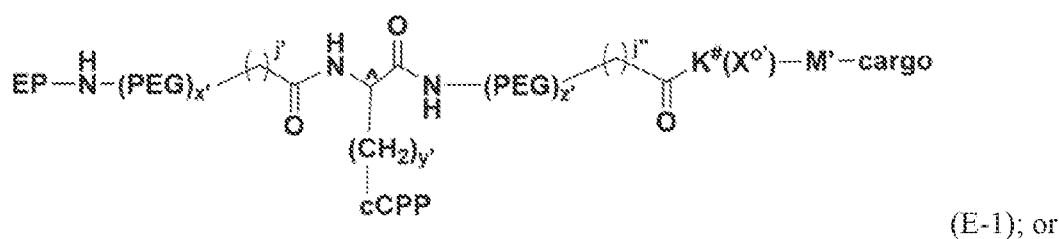
Formula (C-1):



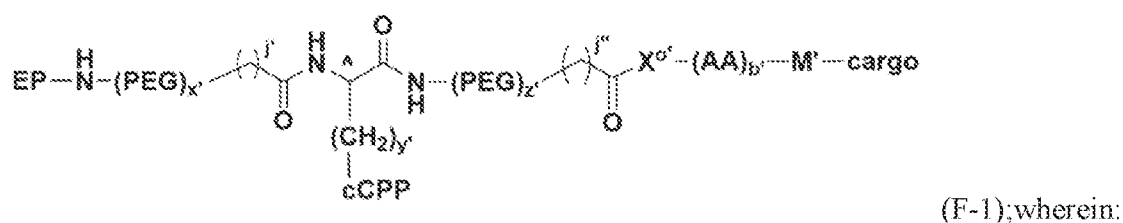
Formula (D-1):



5 Formula (E-1):



Formula (F-1):



EP is a linear exocyclic peptide;

10 cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

15 z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ and $X^{o''}$ are each, independently, a hydrophobic component;

$K^\#$ is a D-lysine or L-lysine residue;

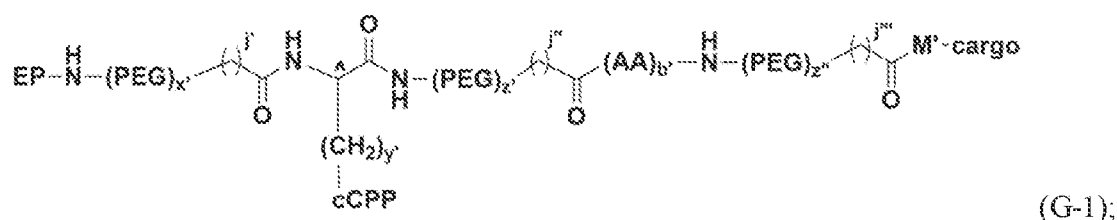
(AA) b' and (AA) b'' are each an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

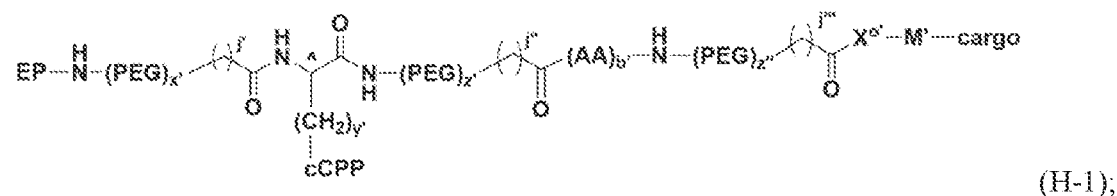
b'' is an integer from 0 to 12.

[0027] In embodiments, the compound of Formula (A-1) comprises a structure selected from:

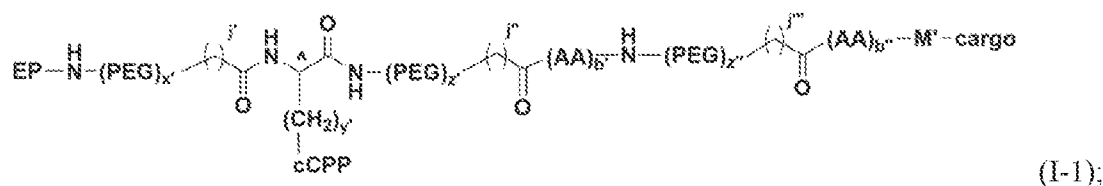
Formula (G-1):



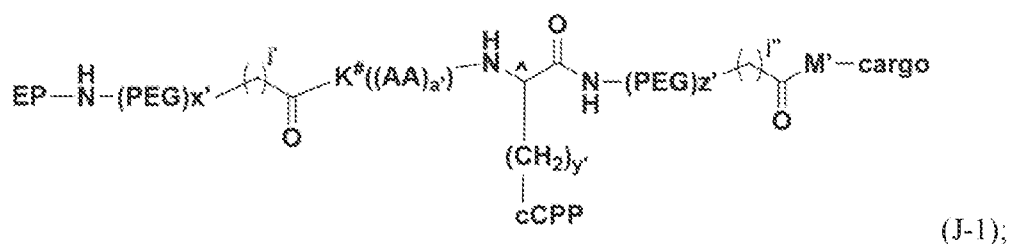
Formula (H-1):



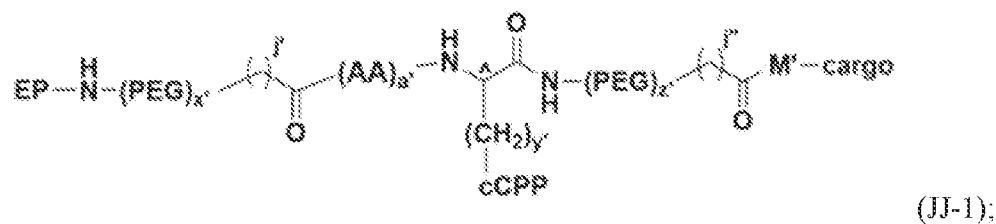
Formula (I-1):



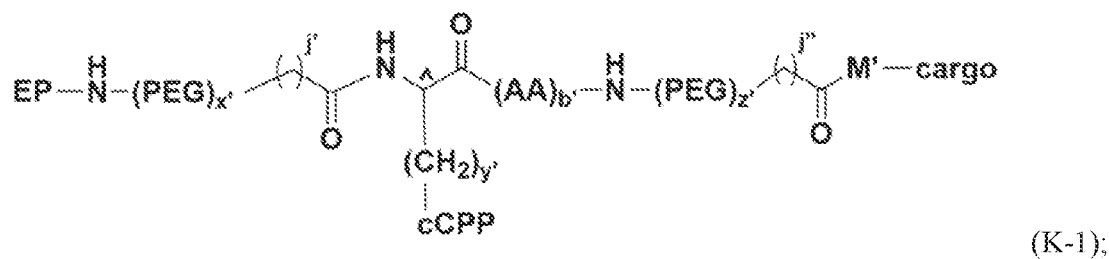
Formula (J-1):



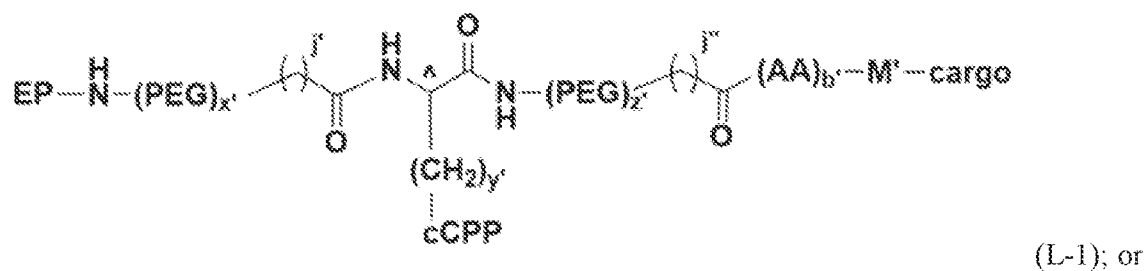
Formula (JJ-1):



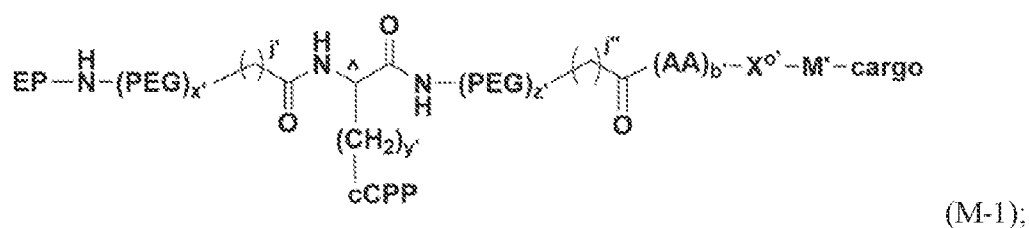
Formula (K-1):



5 Formula (L-1):



Formula (M-1):



wherein:

- 10 EP is a linear exocyclic peptide;
- cCPP is a cyclic cell penetrating peptide;
- x' is an integer from 0 to 12;
- j' is 0, 1, or 2, wherein j' is 0 when x' is 0;
- z' is an integer from 0 to 12;
- 15 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
- z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ comprises a hydrophobic component;

$K^{\#}$ is D-lysine or L-lysine residue;

(AA) a' , (AA) b' , and (AA) b'' are each an amino acid (AA) component comprising at least one amino acid residue;

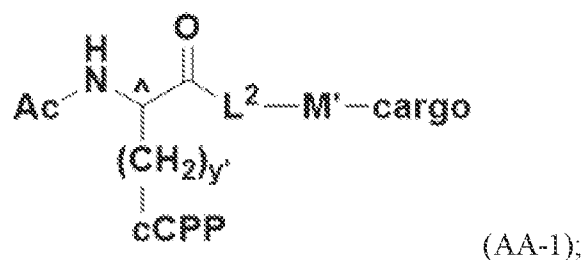
a' is an integer from 0 to 12;

b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

[0028] In embodiments, the EEV can be conjugated to a cargo to form an EEV-cargo conjugate comprising the structure of Formula (AA-1):

Formula (AA-1):



wherein:

cCPP is a cyclic cell penetrating peptide;

Ac is acetyl;

L^1 and L^2 , are, independently, a linker arm;

$\wedge$ indicates L- or D-stereochemistry;

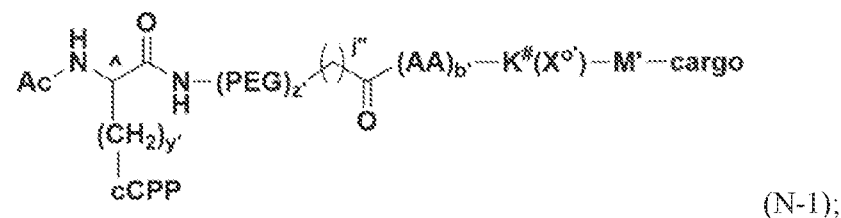
y' is an integer from 1 to 5;

M' comprises a bonding group; and

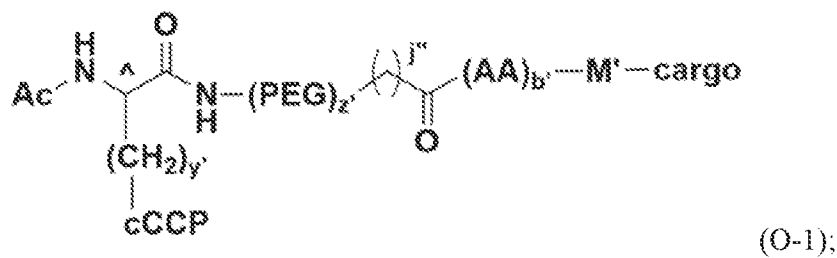
cargo is a peptide, an oligonucleotide, a small molecule, or any combinations thereof.

[0029] In embodiments, the compound of Formula (AA-1) comprises a structure selected from:

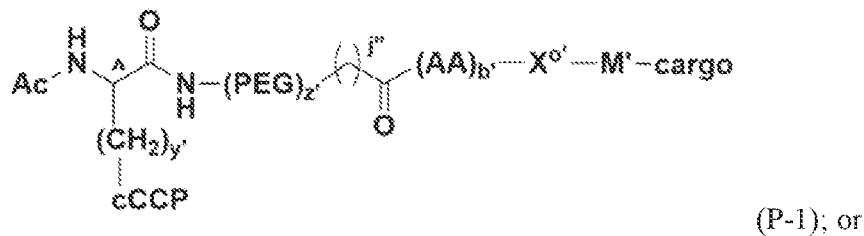
Formula (N-1):



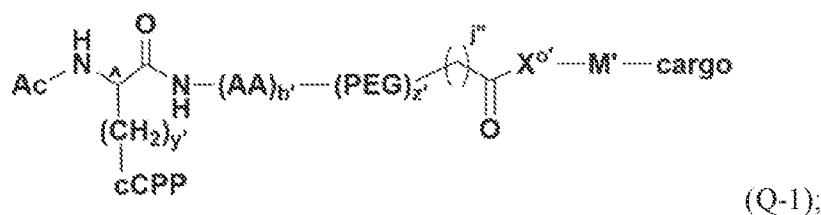
Formula (O-1):



Formula (P-1):



5 *Formula (Q-1):*



wherein,

Ac is acetyl;

cCPP is a cyclic cell penetrating peptide;

10 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^o is a hydrophobic component;

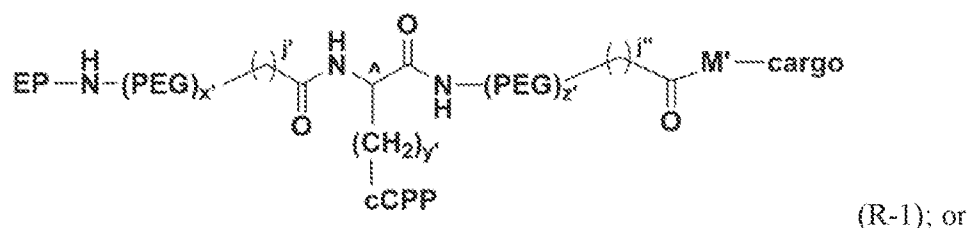
K[#] is D-lysine or L-lysine residue;

(AA)b' is an amino acid (AA) component comprising at least one amino acid residue; and

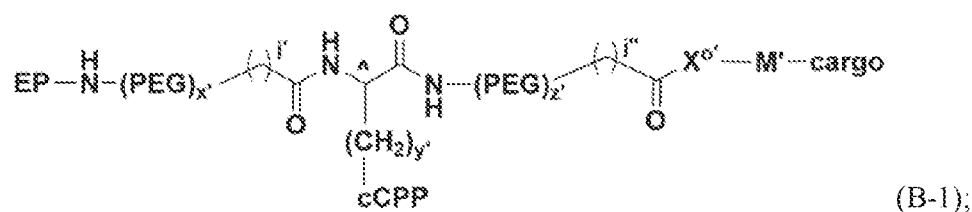
15 b' is an integer from 0 to 12.

[0030] In embodiments, the EEV of Formula (A-1) comprises a structure selected from Formula (R-1) and Formula (B-1):

Formula (R-1):



Formula (B-1):



5 wherein

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

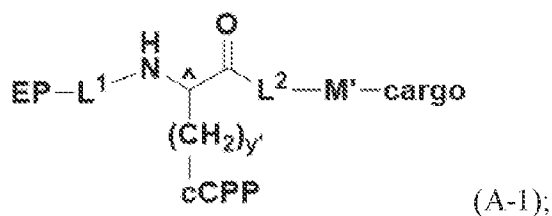
10 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0; and

X^{α'} comprises a hydrophobic component.

[0031] In embodiments, the EEV can be conjugated to a cargo to form an EEV-cargo conjugate comprising the structure of Formula (A-1):

15 Formula (A-1):



wherein

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

20 L¹ and L², are, independently, a linker arm;

^ indicates D-stereochemistry;

y' is an integer from 1 to 5;

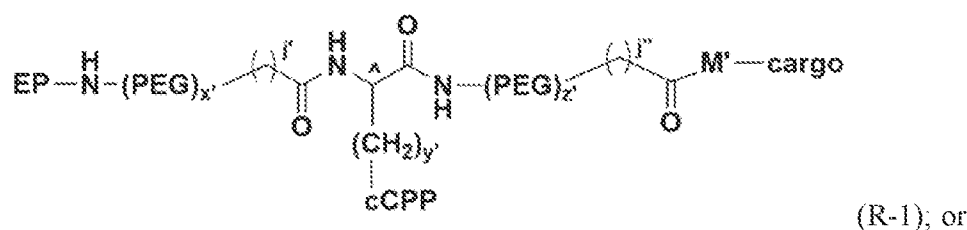
M' comprises a bonding group; and

cargo is a peptide, an oligonucleotide, a small molecule, or any combinations thereof;

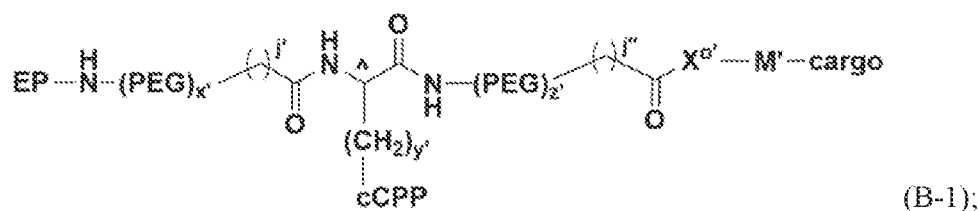
wherein EP comprises all D-amino acids; cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0032] In embodiments, the EEV-cargo conjugate of Formula (A-1) comprises a structure selected from Formula (R-1) and Formula (B-1):

Formula (R-1):



Formula (B-1):



wherein

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$X^{o'}$ comprises a hydrophobic component; and

wherein EP comprises all D-amino acids; cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0033] In embodiments, the EEV-cargo conjugate comprises a structure selected from Formula (B-1), (BB-1), (C-1), (D-1), (E-1), or (F-1), wherein EP comprises all D-amino acids; and cCPP

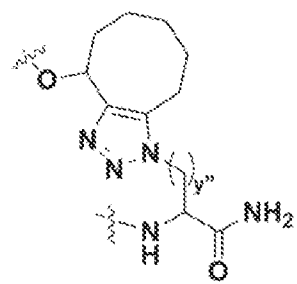
comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0034] In embodiments, the EEV-cargo conjugate comprises a structure selected from Formula (G-1), (H-1), (I-1), (J-1), (JJ-1), (K-1), (L-1), or (M-1), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0035] In embodiments, the EEV-cargo conjugate comprises a structure selected from Formula (N-1), (O-1), (P-1), or (Q-1), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

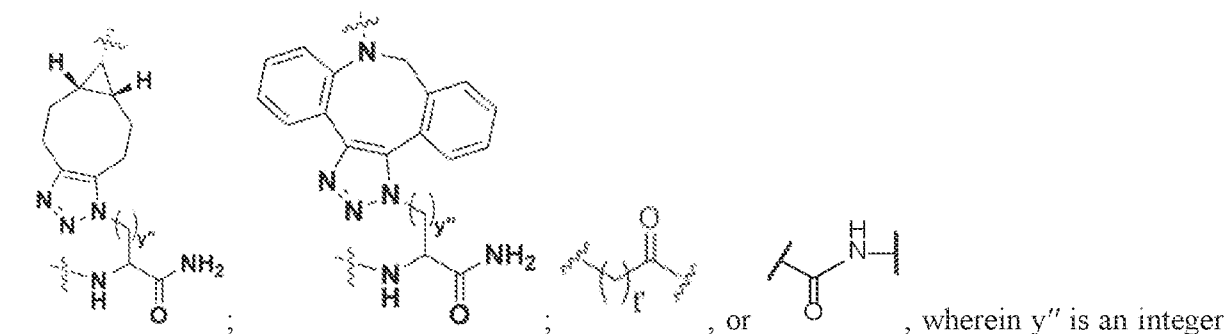
[0036] In embodiments, the EEV-cargo conjugate comprises a structure selected from Formula (R-1) or (B-1), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0037] In embodiments, M' comprises $-NH-$, $-C(O)-$, $-O-$,



from 1 to 4 and t' is an integer from 0 to 10.

[0038] In embodiments, the cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof. In embodiments, the cargo is an oligonucleotide. In embodiments, the oligonucleotide is an antisense oligonucleotide. In embodiments, the oligonucleotide is a phosphorodiamidate morpholino oligonucleotide (PMO).



[0039] In embodiments, the EP comprises from 2 to 10 amino acid residues. In embodiments, the EP comprises from 2 to 8 amino acid residues. In embodiments, the EP comprises from 2 to 6 amino acid residues. In embodiments, the EP comprises 6 amino acid residues. In embodiments, the EP comprises from 7 amino acid residues. In embodiments, the EP comprises from 8 amino acid residues. In embodiments, at least 1 amino acid residue comprises a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof. In embodiments, the EP comprises 1, 2, 3, or 4 lysine residues. In embodiments, the EP comprises 1, 2, 3 or 4 arginine residues. In embodiments, the EP comprises 1 or 2 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises at least 1 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises at least 2 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises 1-5 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises 1-3 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises 1 uncharged hydrophobic amino acid residue. In embodiments, the EP comprises 2 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises 3 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises 4 uncharged hydrophobic amino acid residues. In embodiments, the EP comprises 5 uncharged hydrophobic amino acid residues. In embodiments, the uncharged hydrophobic amino acid residue is selected from valine, proline, β -alanine, glycine, or a combination thereof.

[0040] Also provided herein is a pharmaceutical composition comprising an EEV-cargo conjugate as described herein and a pharmaceutically acceptable carrier and a method of treating a disease in a subject in need thereof comprising administering to the subject an effective amount of the pharmaceutical composition. In embodiments, administering comprises parenteral administration. In embodiments, parenteral administration is selected from: subcutaneous, intradermal, intravenous, intramuscular, intraperitoneal, intrasternal, and intrathecal administration, and intracerebroventricular injection.

BRIEF DESCRIPTION OF THE DRAWINGS

[0041] FIG. 1 shows the effects of linker and conjugation chemistry on efficacy and tolerability.

[0042] FIG. 2 shows the effects of including a hydrophobic component in a linker.

[0043] FIG. 3 shows examples of structural modifications to linkers that mimic the hydrophobic component generated using click chemistry.

[0044] FIG. 4 shows tibialis anterior and heart data on C-terminal Lys-side chain derivatives.

[0045] FIG. 5 shows tibialis anterior and heart data on main chain hydrophobic derivatives.

[0046] FIG. 6 is a structure for EEV Ac-PKKKRKV-PEG₂-K(*cyclo*[FGFGRGRQ])PEG₁₂-(2-Nal)-OH with TFA protecting groups shown on the linear Exocyclic Peptide (EP). It is

understood that the TFA protecting groups can be removed after conjugation to a cargo.

[0047] FIG. 7 is an EEV structure is a structure of EEV Ac-PKKKRKG- PEG₂-

K(*cyclo*[FGFGRGRQ])- PEG₁₂-RB-Bip-RB-OH with TFA protecting groups shown on the linear Exocyclic Peptide (EP). It is understood that the TFA protecting groups can be removed after conjugation to a cargo.

[0048] FIG. 8 is an EEV structure for Ac-PKKKRKG-PEG₂-K(*cyclo*[FGFGRGRQ])-PEG₁₂-Bip-BRBRB-OH with TFA protecting groups shown on the linear Exocyclic Peptide (EP). It is understood that the TFA protecting groups can be removed after conjugation to a cargo.

[0049] FIG. 9 is an EEV structure for Ac-PKKKRKV-PEG₂-K(*cyclo*[FfFGRGRQ])-PEG₂-RF-PEG₄-OH with TFA protecting groups shown on the linear Exocyclic Peptide (EP). It is

understood that the TFA protecting groups can be removed after conjugation to a cargo.

[0050] FIG. 10 is an EEV structure for Ac-PKKKRKV-PEG₂-K(*cyclo*[FGFGRGRQ])-PEG₁₂-Nal-PEG₂-Nal-COOH with TFA protecting groups shown on the linear Exocyclic Peptide (EP). It is understood that the TFA protecting groups can be removed after conjugation to a cargo.

[0051] FIG. 11 is an EEV structure for Ac-PKKKRKV-PEG₂-K(*cyclo*[FGFGRGRQ])-PEG₁₂-

K(Ac-Nal)-OH with TFA protecting groups shown on the linear Exocyclic Peptide (EP). It is understood that the TFA protecting groups can be removed after conjugation to a cargo.

[0052] FIG. 12 is an EEV structure for Ac-KKKRK-PEG₂-K(RBRRBR-Ac)-K(*cyclo*[FGFGRGRQ])-PEG₁₂-K(N₃)-NH₂.

DETAILED DESCRIPTION

Endosomal Escape Vehicles (EEVs)

[0053] An endosomal escape vehicle (EEV) is provided herein that can be used to transport a cargo molecule across a cellular membrane, for example, to deliver the cargo to the cytosol or nucleus of a cell. The cargo can be a therapeutic or diagnostic molecule. The therapeutic molecule can be an oligonucleotide, peptide, small molecule or a combination thereof. The peptide can be a protein, an antibody or an enzyme. The cargo can be an oligonucleotide. The oligonucleotide can be an

antisense oligonucleotide. The oligonucleotide can be a phosphorodiamidate morpholino oligonucleotide (PMO). The EEV can comprise a cell penetrating peptide (CPP), for example, a cyclic cell penetrating peptide (cCPP), which is conjugated to a linear exocyclic peptide (EP). The EEV can include one or more linkers. The EEV can include a hydrophobic component (X). One or more linker arms of the EEV or the cCPP can include a hydrophobic component (X). In embodiments, when a linker arm of the EEV includes more than one hydrophobic component (X), the hydrophobic components can be indicated using (e.g., X^{o'}, X^{o''}, etc.). In embodiments, the EEV can comprise hydrocarbon component (e.g., NRH-(CH₂)_n-COOH), a polyethylene glycol (PEG) component, a hydrophobic component (X), an amino acid component comprising one or more amino acid residues (AA), or a combination thereof. In embodiments, one or more components comprise (AA), (PEG), a hydrophobic component (X) or a combination thereof. In embodiments, EEV constructs in which the stereochemistry of the amino acids within the peptide sequences of the construct has been modified are provided.

[0054] In embodiments, the EEV is coupled to a cargo. The cargo can be coupled to the cCPP.

The cargo can be coupled to the EP. The cargo can be coupled to a linker. The EP can be coupled to the cCPP. The EP can be coupled to the cargo and the cCPP. Coupling between the EP, cargo, cCPP, or combinations thereof, may be non-covalent or covalent. The EP, cargo, cCPP, or combination thereof, may be coupled via one or more linkers.

Cell Penetrating Peptides (CPP)

[0055] A delivery construct is provided herein that comprises at least one cell penetrating peptide (CPP). The cell penetrating peptide can be a cyclic cell penetrating peptide (cCPP). In embodiments, the cCPP can penetrate a cell membrane. In embodiments, the cCPP can deliver the cargo to the cytosol of the cell. The cCPP can deliver the cargo to a cellular location where a target gene, target transcript, and/or target protein is located. To conjugate the cCPP to a cargo, an EP, and/or a linker, at least one bond or lone pair of electrons on the cCPP can be replaced.

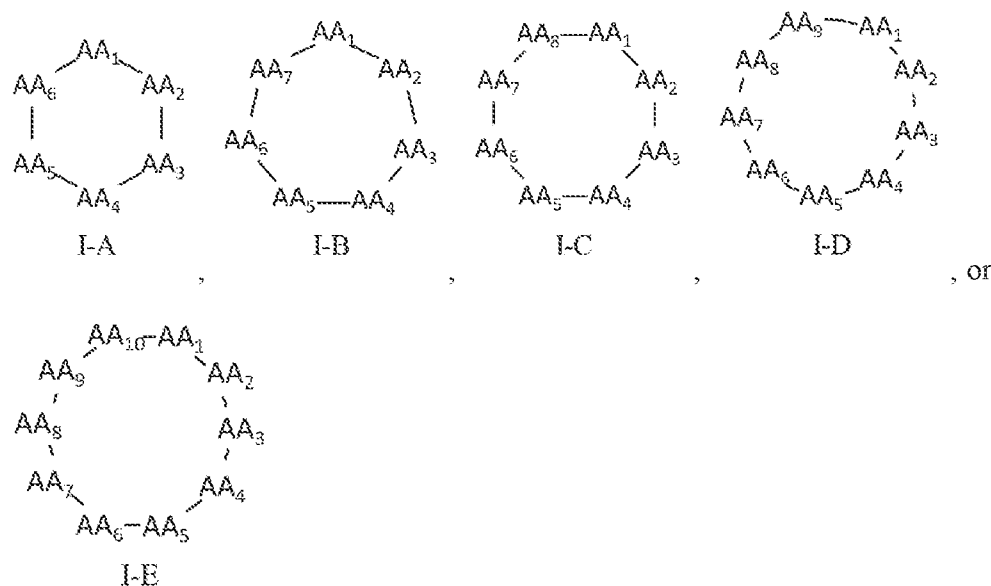
[0056] In embodiments, a hydrophobic component (X) is appended to one or more amino acid residues of the cCPP. In embodiments, a hydrophobic component (X) is appended to a side chain of a lysine amino acid residue of the cCPP.

[0057] X can be a D or L amino acid residue with a hydrophobic side chain. X can be a naturally occurring or a non-naturally occurring amino acid residue with a hydrophobic side chain. X can be an amino acid residue with an aromatic side chain. X can be an amino acid residue with a

heteroaromatic side chain. X can be selected from phenylalanine, 3-(4',4-biphenyl)-L-alanine, tryptophan, tyrosine, valine, isoleucine, leucine, or histidine, or a combination thereof. X can be 2-naphthylalanine. X can be Nal. X can be d-Nal (nal). X can be 3-(4',4-biphenyl)-L-alanine. X can be Bip. X can be D-Bip (bip). X can be a C₄-C₈ alkyl hydrocarbon. X can be a C₆ alkyl hydrocarbon.

[0058] The total number of amino acid residues in the cCPP is from 6 to 20 amino acid residues, e.g., 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, or 20 amino acid residues, inclusive of all ranges and subranges therebetween. The cCPP can comprise from 6 to 13 amino acid residues. The cCPP can comprise from 6 to 10 amino acids. By way of example, cCPP comprising 6-10 amino acid residues can have a structure according to any of Formula I-A to I-E:

[0059] Formula I-A to I-E:



, wherein AA₁, AA₂, AA₃, AA₄, AA₅, AA₆, AA₇, AA₈, AA₉, and AA₁₀ are amino acid residues.

[0060] The cCPP can comprise from 6 to 8 amino acids. The cCPP can comprise 8 amino acids.

[0061] Each amino acid in the cCPP may be a natural or non-natural amino acid. Abbreviations used herein for some natural and non-natural amino acids are shown in **Table 1**.

[0062] As used herein, the term "amino acid" refers to compounds having an amino group and a carboxylic acid group. Most amino acids (except for glycine) also have a side chain. As used herein, "amino acid side chain" or "side chain" refers to the characterizing substituent bound to the α-carbon of the amino acid.

[0063] An “ α -amino acid” is an amino acid in which the amino group is attached to the first (alpha) carbon adjacent to the carboxylic acid group, such that the carbon atom of the carbonyl is separated from the nitrogen atom of the amino group by one carbon atom. A “ β -amino acid” (also called “beta-amino acid,” and “ β -amino acid”) is an analog of an α -amino acid in which the amino group is attached to the second (beta) carbon, rather than the alpha-carbon, such that the carbon atom of the carbonyl is separated from the nitrogen atom of the amino group by two carbon atoms. Examples of β -amino acids include but are not limited to β -alanine and β -homophenylalanine.

[0064] An “uncharged” amino acid is an amino acid that does not have a charge at a physiological pH (for example, from 6.5 to 8.0 or from 6.8 to 7.6). It is noted that histidine can exist in neutral or positively charged forms at physiological pH.

[0065] A side chain that does not comprise an aryl or heteroaryl group, can be referred to herein as a “non-aryl” side chain. In embodiments, the side chain that does not comprise an aryl or heteroaryl group can be uncharged and is referred to herein as an uncharged, non-aryl side chain. Amino acids with uncharged non-aryl amino side chains include, but are not limited to, histidine, threonine, serine, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, 3-(4-thienyl)-alanine, and β -amino acid derivatives thereof.

[0066] The term “non-natural amino acid” refers to an organic compound that is a congener of a natural amino acid in that it has a structure similar to a natural amino acid so that it mimics the structure and reactivity of a natural amino acid. The non-natural amino acid can be a modified amino acid, and/or amino acid analog, that is not one of the 20 common naturally occurring amino acids or the rare natural amino acids selenocysteine or pyrrolysine. Non-natural amino acids can also be a D-isomer of a natural amino acid. Examples of suitable amino acids include, but are not limited to, alanine, allosoleucine, arginine, citrulline, asparagine, aspartic acid, cysteine, glutamine, glutamic acid, glycine, histidine, isoleucine, leucine, lysine, methionine, naphthylalanine, phenylalanine, proline, pyroglutamic acid, serine, threonine, tryptophan, tyrosine, valine, a derivative thereof, or combinations thereof.

Table 1: Amino Acid Abbreviations

Amino Acid	Abbreviations* L-amino acid	Abbreviations* D-amino acid
Alanine	Ala (A)	ala (a)
Allo-isoleucine	Aile	aile

Amino Acid	Abbreviations* L-amino acid	Abbreviations* D-amino acid
Arginine	Arg (R)	arg (r)
Asparagine	Asn (N)	asn (n)
Aspartic acid	Asp (D)	asp (d)
Cysteine	Cys (C)	cys (c)
Citrulline	Cit	cit
Cyclohexylalanine	Cha	cha
2,3-diaminopropionic acid	Dap	dap
4-fluorophenylalanine	Fpa (Σ)	pfa
Glutamic acid	Glu (E)	glu (e)
Glutamine	Gln (Q)	gln (q)
Glycine	Gly (G)	gly (g)
Histidine	His (H)	his (h)
Homoproline (aka pipecolic acid)	Pip (Θ)	pip (θ)
Isoleucine	Ile (I)	ile (i)
Leucine	Leu (L)	leu (l)
Lysine	Lys (K)	lys (k)
Methionine	Met (M)	met (m)
3-(2-naphthyl)-alanine	Nal (Φ) (L-Nal or 2-Nal)	nal (ϕ) (dNal, dnal, D-Nal, or 2-nal)
3-(1-naphthyl)-alanine	1-Nal	1-nal
Norleucine	Nle (Ω)	nle
Phenylalanine	Phe (F)	phe (f)
Phenylglycine	Phg (Ψ)	phg
4-(phosphonodifluoromethyl)phenylalanine	F ₂ Pmp (Λ)	f ₂ pmp
Proline	Pro (P)	pro (p)
Sarcosine	Sar (Ξ)	sar
Selenocysteine	Sec (U)	sec (u)
Serine	Ser (S)	ser (s)
Threonine	Thr (T)	thr (y)
Tyrosine	Tyr (Y)	tyr (y)
Tryptophan	Trp (W)	trp (w)
Valine	Val (V)	val (v)
Tert-butyl-alanine	Tle	tle
Penicillamine	Pen	pen
Homoarginine	HomoArg	homoarg
Nicotinyl-lysine	Lys(NIC)	lys(NIC)
Trifluoroacetyl-lysine	Lys(TFA)	lys(TFA)
Methyl-leucine	MeLeu	meleu
3-(3-benzothieryl)-alanine	Bta	bta
* single letter abbreviations: capital letters indicate the L-amino acid form, lower case letter indicate the D-amino acid form.		

[0067] In embodiments, the cCPP can comprise 2 contiguous amino acids with hydrophobic side chains. In embodiments, the cCPP can comprise 3 contiguous amino acids with hydrophobic side chains. In embodiments, the amino acids with hydrophobic side chain are selected from phenylalanine and naphthylalanine. In embodiments, the contiguous amino acids with hydrophobic side chains can have D- or L-stereochemistry. In embodiments, the contiguous amino acids with hydrophobic side chains can have the same stereochemistry. In embodiments, the contiguous amino acids with hydrophobic side chains can have alternating stereochemistry. In embodiments, the contiguous amino acids with hydrophobic side chains can have all L-stereochemistry. In embodiments, the contiguous amino acids with hydrophobic side chains can have all D-stereochemistry.

[0068] In embodiments, one or two amino acids in the cCPP can have no side chain. In embodiments, all amino acids in the cCPP have a side chain. As used herein, when no side chain is present, the amino acid has two hydrogen atoms on the carbon atom(s) (e.g., -CH₂) linking the amine and carboxylic acid of the amino acid residue. The amino acid having no side chain can be glycine. The amino acid having no side chain can be β -alanine.

[0069] In embodiments, the cCPP can comprise 2 contiguous amino acids either hydrophobic side chains, no side chain, or a combination thereof. In embodiments, the cCPP can comprise 3 contiguous amino acids with hydrophobic side chains, no side chain, or a combination thereof. In embodiments, the contiguous amino acids with hydrophobic side chains, no side chains, or combination thereof can have alternating stereochemistry. In embodiments, the contiguous amino acids with hydrophobic side chains, no side chains, or combination thereof can have all L-stereochemistry. In embodiments, the contiguous amino acids with hydrophobic side chains, no side chains, or combination thereof can have all D-stereochemistry.

[0070] The cCPP can comprise from 6 to 20, from 6 to 10, or from 6 to 8 amino acid residues, wherein: (i) at least two amino acids can, independently, be glycine, β -alanine, serine, histidine, citrulline, or 4-aminobutyric acid; (ii) at least two amino acids can have a side chain comprising an aryl or heteroaryl group; and (iii) at least two amino acids, independently, have a side chain comprising a guanidine group, or a protonated form thereof. In embodiments, (i) two amino acids can, independently, be glycine, β -alanine, serine, histidine, citrulline, or 4-aminobutyric acid; (ii) two or three amino acids can have a side chain comprising an aryl or heteroaryl group; and (iii)

two amino acids, independently, have a side chain comprising a guanidine group, or a protonated form thereof.

[0071] In embodiments, one amino acid of the cCPP can be glycine, β -alanine, serine, histidine, citrulline, or 4-aminobutyric acid. In embodiments, two amino acids can be, independently, glycine, β -alanine, serine, histidine, citrulline, or 4-aminobutyric acid. In embodiments, three amino acids can be glycine, β -alanine, serine, histidine, citrulline, or 4-aminobutyric acid.

[0072] In embodiments, one amino acid of the cCPP can have a side chain comprising an aryl or heteroaryl group. In embodiments, two amino acids of the cCPP can have a side chain comprising an aryl or heteroaryl group. In embodiments, three amino acids of the cCPP can have a side chain comprising an aryl or heteroaryl group. In embodiments, none of the amino acids having the side chain comprising the aryl or heteroaryl group are contiguous. In embodiments, two amino acids having the side chain comprising the aryl or heteroaryl group can be contiguous. In embodiments, two contiguous amino acids can have opposite stereochemistry. In embodiments, the two contiguous amino acids can have the same stereochemistry. In embodiments, three amino acids having the side chain comprising the aryl or heteroaryl group can be contiguous. In embodiments, three contiguous amino acids can have the same stereochemistry. In embodiments, three contiguous amino acids can have alternating stereochemistry. The amino acid residue having a side chain comprising an aryl or heteroaryl group can each be independently a residue of phenylalanine, naphthylalanine, or β -homophenylalanine, each of which is optionally substituted with one or more substituents. At least one amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of phenylalanine. One amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of phenylalanine. At least two amino acid residues having a side chain comprising an aryl or heteroaryl group can be residues of phenylalanine. Two amino acid residues having a side chain comprising an aryl or heteroaryl group can be residues of phenylalanine. Each amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of phenylalanine. At least one amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of naphthylalanine. One amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of naphthylalanine. At least one amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of β -homophenylalanine. One amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of β -homophenylalanine.

[0073] In embodiments, one amino acid of the cCPP can have a side chain that does not comprise an aryl or heteroaryl group, referred to herein as a “non-aryl” side chain. In embodiments, the side chain that does not comprise an aryl or heteroaryl group can be uncharged and is referred to herein as an uncharged, non-aryl side chain. In embodiments, two amino acids of the cCPP can have an uncharged, non-aryl side chain. In embodiments, three amino acids of the cCPP can have an uncharged, non-aryl side chain. Amino acids with uncharged non-aryl amino side chains include, but are not limited to, histidine, threonine, serine, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine.

[0074] In embodiments, one amino acid of the cCPP has a side chain comprising a guanidine group, or a protonated form thereof. In embodiments, two amino acids of the cCPP can have a side chain comprising a guanidine group, or a protonated form thereof. In embodiments, three amino acids of the cCPP can have a side chain comprising a guanidine group, or a protonated form thereof. In embodiments, four amino acids of the cCPP can have a side chain comprising a guanidine group, or a protonated form thereof. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof are contiguous. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof are not contiguous. In embodiments, the amino acid comprising a guanidine group or protonated form thereof is arginine. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof have D- or L- stereochemistry. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof have alternating D- and L- stereochemistry. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof have the same stereochemistry. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof have D- stereochemistry. In embodiments, the amino acids of the cCPP having a side chain comprising a guanidine group, or a protonated form thereof have L- stereochemistry.

[0075] In embodiments, the cCPP can comprise from 6 to 10 amino acid residues, wherein: (i) at least two amino acids can, independently, be glycine, β -alanine, or 4-aminobutyric acid residues; (ii) at least two amino acids can have a side chain comprising an aryl or heteroaryl group; and (iii)

at least two amino acids can, independently, have a side chain comprising a guanidine group, or a protonated form thereof.

[0076] In embodiments, the cCPP can comprise from 6 to 10 amino acid residues, wherein: (i) two amino acid can independently be glycine, β -alanine, or 4-aminobutyric acid residues; (ii) two or
5 three amino acids can have a side chain comprising an aryl or heteroaryl group; and (iii) two amino acid can, independently, have a side chain comprising a guanidine group, or a protonated form thereof.

[0077] In embodiments, the cCPP can comprise from 6 to 10 amino acid residues, wherein: (i) at least two amino acids can independently be glycine, β -alanine, or 4-aminobutyric acid residues;
10 (ii) at least three amino acids can have a side chain comprising an aryl or heteroaryl group; and (iii) at least two amino acids comprise arginine.

Glycine and Related Amino Acid Residues

[0078] In embodiments, the cCPP can comprise 1, 2, 3, 4, 5, or 6 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 2
15 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 3 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 4 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 5 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 6
20 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 3, 4, 5, or 6 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 3, 4, or 5 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof. In embodiments, the cCPP can comprise 3 or 4 glycine, β -alanine, 4-aminobutyric acid residues, or combinations thereof.

[0079] In embodiments, the cCPP can comprise 1, 2, 3, 4, 5, or 6 glycine residues. The cCPP can comprise 2 glycine residues. In embodiments, the cCPP can comprise 3 glycine residues. In
25 embodiments, the cCPP can comprise 4 glycine residues. In embodiments, the cCPP can comprise 5 glycine residues. In embodiments, the cCPP can comprise 6 glycine residues. In embodiments, the cCPP can comprise 3, 4, or 5 glycine residues. In embodiments, the cCPP can comprise 3 or 4

glycine residues. In embodiments, the cCPP can comprise 2 or 3 glycine residues. In embodiments, the cCPP can comprise 1 or 2 glycine residues.

[0080] In embodiments, the cCPP can comprise at least three glycine residues. In embodiments, the cCPP can comprise 3, 4, 5, or 6 glycine residues. In embodiments, the cCPP can comprise 3 glycine residues. In embodiments, the cCPP can comprise 4 glycine residues. In embodiments, the cCPP can comprise 5 glycine residues. In embodiments, the cCPP can comprise 6 glycine residues. In embodiments, the cCPP can comprise 3, 4, or 5 glycine residues. The cCPP can comprise 3 or 4 glycine residues.

[0081] In embodiments, none of the glycine, β -alanine, or 4-aminobutyric acid residues in the cCPP are contiguous. In embodiments, two or three glycine, β -alanine, 4-or aminobutyric acid residues can be contiguous. In embodiments, two glycine, β -alanine, or 4-aminobutyric acid residues can be contiguous.

[0082] In embodiments, none of the glycine residues in the cCPP are contiguous. For example, each glycine residues in the cCPP can be separated by an amino acid residue that is not glycine.

[0083] In embodiments, two or more of the glycine residues in the cCPP are contiguous. In embodiments, two or three glycine residues can be contiguous. In embodiments, two glycine residues are contiguous.

Amino Acid Side Chains with an Aryl or heteroaryl Group

[0084] In embodiments, the cCPP can comprise 2, 3, 4, 5, or 6 amino acid residues independently having a side chain comprising an aryl or heteroaryl group. In embodiments, the cCPP can comprise 2 amino acid residues independently having a side chain comprising an aryl or heteroaryl group. In embodiments, the cCPP can comprise 3 amino acid residues independently having a side chain comprising an aryl or heteroaryl group. In embodiments, the cCPP can comprise 2, 3, or 4 amino acid residues independently having a side chain comprising an aryl or heteroaryl group. In embodiments, the cCPP can comprise 2 or 3 amino acid residues independently having a side chain comprising an aryl or heteroaryl group.

[0085] The aryl group can be a 6- to 14-membered aryl. Aryl can be phenyl, naphthyl or anthracenyl, each of which is optionally substituted. Aryl can be phenyl or naphthyl, each of which is optionally substituted. The heteroaryl group can be a 6- to 14-membered heteroaryl having 1, 2, or 3 heteroatoms selected from N, O, and S. Heteroaryl can be pyridyl, quinolyl, or isoquinolyl.

[0086] The amino acid residue having a side chain comprising an aryl or heteroaryl group can each be independently a residue of phenylalanine, naphthylalanine, phenylglycine, β -homophenylalanine, homonaphthylalanine, bis(homophenylalanine), bis-(homonaphthylalanine), tryptophan, or tyrosine, each of which is optionally substituted with one or more substituents. The

5 amino acid residue having a side chain comprising an aryl or heteroaryl group can each independently be a residue of tyrosine, phenylalanine, 1-naphthylalanine, 2-naphthylalanine, tryptophan, 3-benzothienylalanine, 4-phenylphenylalanine, 3,4-difluorophenylalanine, 4-trifluoromethylphenylalanine, 2,3,4,5,6-pentafluorophenylalanine, homophenylalanine, β -homophenylalanine, 4-tert-butyl-phenylalanine, 4-pyridinylalanine, 3-pyridinylalanine, 4-methylphenylalanine, 4-fluorophenylalanine, 4-chlorophenylalanine, 3-(9-anthryl)-alanine. The

10 amino acid residue having a side chain comprising an aryl or heteroaryl group can each independently be a residue of 3-(3-benzothienyl)-alanine, 3-(2-quinolyl)-alanine, O-benzylserine, 3-(4-(benzyloxy)phenyl)-alanine, S-(4-methylbenzyl)cysteine, *N*-(naphthalen-2-yl)glutamine, 3-(1,1'-biphenyl-4-yl)-alanine, 3-(3-benzothienyl)-alanine. The amino acid residue having a side

15 chain comprising an aryl or heteroaryl group can each independently be a residue of phenylalanine, 1-naphthylalanine, 2-naphthylalanine, phenylglycine, β -homophenylalanine, or homonaphthylalanine, each of which is optionally substituted with one or more substituents. The

amino acid residue having a side chain comprising an aryl or heteroaryl group can each be independently a residue of phenylalanine, 2-naphthylalanine, or β -homophenylalanine, each of

20 which is optionally substituted with one or more substituents. The amino acid residue having a side chain comprising an aryl or heteroaryl group can each be independently a residue of phenylalanine or 2-naphthylalanine, each of which is optionally substituted with one or more substituents. At least one amino acid residue having a side chain comprising an aryl or heteroaryl

group can be a residue of phenylalanine. At least two amino acid residues having a side chain

25 comprising an aryl or heteroaryl group can be residues of phenylalanine. Each amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of phenylalanine. One

amino acid residue having a side chain comprising an aryl or heteroaryl group can be a residue of 2-naphthylalanine. One amino acid residue having a side chain comprising an aryl or heteroaryl

group can be a residue of β -homophenylalanine

30 [0087] In embodiments, none of the amino acids having the side chain comprising the aryl or heteroaryl group are contiguous. In embodiments, two amino acids having the side chain

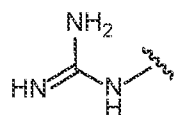
comprising the aryl or heteroaryl group can be contiguous. In embodiments, two contiguous amino acids can have opposite stereochemistry. In embodiments, the two contiguous amino acids have the same stereochemistry. In embodiments, three amino acids having the side chain comprising the aryl or heteroaryl group can be contiguous. In embodiments, three contiguous amino acids having a side chain comprising an aryl or heteroaryl group have the same stereochemistry. In embodiments, three contiguous amino acids have alternating stereochemistry.

[0088] The amino acid residues comprising aryl or heteroaryl groups can be L-amino acids. The amino acid residues comprising aryl or heteroaryl groups can be D-amino acids. The amino acid residues comprising aryl or heteroaryl groups can be a mixture of D- and L-amino acids.

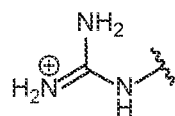
[0089] The hydrophobicity of amino acid residues can be measured and/or calculated using a variety of techniques. In embodiments, the hydrophobicity of an amino acid residue can be determined by calculating its consensus value on the consensus scale of D. Eisenberg et al., using the method described in D. Eisenberg et al., "Hydrophobic Moments and Protein Structure," Faraday Symp. Chem. Soc. 1982, 17, 109-120 (e.g., D. Eisenberg et al.). A hydrophobic amino acid is an amino acid that has a hydrophobic side chain.

Amino Acid Residues Having a Side Chain Comprising a Guanidine Group, Guanidine Replacement Group, or Protonated Form Thereof

[0090] As used herein, guanidine refers to the structure:



[0091] As used herein, a protonated form of guanidine refers to the structure:



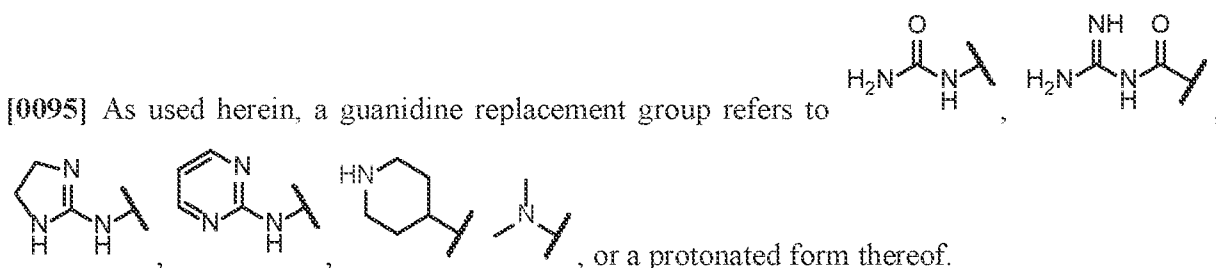
[0092] Guanidine replacement groups refer to functional groups on the side chain of amino acids that will be positively charged at or above physiological pH or those that can recapitulate the hydrogen bond donating and accepting activity of guanidinium groups.

[0093] While not wishing to be bound by theory, it is believed that guanidine replacement groups may facilitate cell penetration and delivery of a therapeutic agent while reducing toxicity associated with guanidine groups or protonated forms thereof. The cCPP can comprise at least one

amino acid having a side chain comprising a guanidine or guanidinium replacement group. The cCPP can comprise two amino acids having a side chain comprising a guanidine or guanidinium replacement group. The cCPP can comprise three amino acids having a side chain comprising a guanidine or guanidinium replacement group. The cCPP can comprise four amino acids having a side chain comprising a guanidine or guanidinium replacement group. The cCPP can comprise five amino acids having a side chain comprising a guanidine or guanidinium replacement group. The cCPP can comprise six amino acids having a side chain comprising a guanidine or guanidinium replacement group.

[0094] The guanidine or guanidinium group can be an isostere of guanidine or guanidinium. The guanidine or guanidinium replacement group can be less basic than guanidine.

[0095] As used herein, a guanidine replacement group refers to



[0096] In embodiments, a cCPP comprising from 6 to 10 amino acids residues is provided, wherein: (i) at least two amino acid have a side chain comprising a guanidine group, or a protonated form thereof; (ii) at least one amino acid residue has no side chain or a side chain of a guanidine replacement group, or a protonated form thereof; and (iii) at least two amino acids residues independently have a side chain comprising an aryl or heteroaryl group. In embodiments, two amino acids independently have a side chain comprising an aryl or heteroaryl group. In embodiments, three amino acids independently have a side chain comprising an aryl or heteroaryl group. In embodiments, at least two amino acids have no side chain or a side chain comprising a guanidine replacement group or a protonated form thereof. As used herein, when no side chain is present, the amino acid has two hydrogen atoms on the carbon atom(s) (e.g., -CH₂-) linking the amine and carboxylic acid. In embodiments, the amino acid having no side chain can be glycine.

[0097] In embodiments, the cCPP can comprise 2, 3, 4, 5, or 6 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 2 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 3 amino acid residues independently having a

side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 4 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 5 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 6 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 2, 3, 4, or 5 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 2, 3, or 4 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof. In embodiments, the cCPP can comprise 2 or 3 amino acid residues independently having a side chain comprising a guanidine group, guanidine replacement group, or a protonated form thereof.

[0098] In embodiments, the amino acid residues independently having the side chain comprising the guanidine group, guanidine replacement group, or the protonated form thereof, can be L-amino acids. In embodiments, the amino acid residues independently having the side chain comprising the guanidine group, guanidine replacement group, or the protonated form thereof, can be D-amino acids. In embodiments, the amino acid residues independently having the side chain comprising the guanidine group, guanidine replacement group, or the protonated form thereof, can be a mixture of L- or D-amino acids.

[0099] In embodiments, each amino acid residue having the side chain comprising the guanidine group, or the protonated form thereof, can independently be a residue of arginine, homoarginine, 2-amino-3-propionic acid, 2-amino-4-guanidinobutyric acid or a protonated form thereof. In embodiments, each amino acid residue having the side chain comprising the guanidine group, or the protonated form thereof, can independently be a residue of arginine or a protonated form thereof. In embodiments, the amino acid residue having the side chain comprising the guanidine replacement group, or the protonated form thereof, can independently be a residue of citrulline.

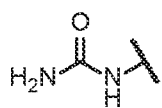
[0100] In embodiments, the cCPP can comprise a residue of asparagine, aspartic acid, glutamine, glutamic acid, or homoglutamine. In embodiments, the cCPP can comprise a residue of asparagine. In embodiments, the cCPP can comprise a residue of glutamine.

Chirality

[0101] While not wishing to be bound by theory, it is believed that the chirality of the amino acids in the cCPPs may impact cytosolic uptake efficiency. In embodiments, the cCPP can comprise at least one D amino acid. In embodiments, the cCPP can comprise 1 to 20 D amino acids. In
5 embodiments, the cCPP can comprise 1 to 15 D amino acids. In embodiments, the cCPP can comprise 1 to 10 D amino acids. In embodiments, the cCPP can comprise 1 to 8 D amino acids. In
embodiments, the cCPP can comprise 1, 2, 3, 4, 5, 6, 7, or 8 D amino acids. In embodiments, the
cCPP can comprise all D amino acids. In embodiments, the cCPP can comprise at least one L
amino acid. In embodiments, the cCPP can comprise 1 to 20 L amino acids. In embodiments, the
10 cCPP can comprise 1 to 15 L amino acids. In embodiments, the cCPP can comprise 1 to 10 L
amino acids. In embodiments, the cCPP can comprise 1 to 8 L amino acids. In embodiments, the
cCPP can comprise 1, 2, 3, 4, 5, 6, 7, or 8 L amino acids. In embodiments, the cCPP can comprise
all L amino acids. In embodiments, the cCPP can comprise 2, 3, 4, 5, 6, 7, or 8 contiguous amino
15 acids having alternating D and L chirality. In embodiments, the cCPP can comprise three
contiguous amino acids having the same chirality. In embodiments, the cCPP can comprise two
contiguous amino acids having the same chirality. In embodiments, at least two of the amino acids
can have the opposite chirality. In embodiments, at least two amino acids having opposite chirality
can be adjacent to each other. In embodiments, at least three amino acids can have alternating
20 stereochemistry relative to each other. In embodiments, the at least three amino acids having the
alternating chirality relative to each other can be adjacent to each other. In embodiments, at least
four amino acids have alternating stereochemistry relative to each other. In embodiments, the at
least four amino acids having the alternating chirality relative to each other can be adjacent to each
other. In embodiments, at least two of the amino acids can have the same chirality. In
embodiments, at least two amino acids having the same chirality can be adjacent to each other. In
25 embodiments, at least two amino acids have the same chirality and at least two amino acids have
the opposite chirality. In embodiments, the at least two amino acids having the opposite chirality
can be adjacent to the at least two amino acids having the same chirality. Accordingly, adjacent
amino acids in the cCPP can have any of the following sequences: D-L; L-D; D-L-D, L-D-L, D-
L-L-D; L-D-D-L; L-D-L-L-D; D-L-D-D-L; D-L-L-D-L; or L-D-D-L-D. In embodiments, the
30 amino acid residues that form the cCPP can all be L-amino acids. In embodiments, the amino acid
residues that form the cCPP can all be D-amino acids.

[0102] In embodiments, one or more amino acid residues that form the cCPP can be achiral. In embodiments, the cCPP can comprise a motif of 3, 4, or 5 amino acids, wherein two amino acids having the same chirality can be separated by an achiral amino acid. The cCPPs can comprise the following sequences: D/L-X-D/L; D/L-X-D/L-X; D/L-X-D/L-X-D/L; D-X-D; D-X-D-X; D-X-D-X-D; L-X-L; L-X-L-X; or L-X-L-X-L, wherein D/L indicates that the amino acid can be a D or an L amino acid and X is an achiral amino acid. The achiral amino acid can be glycine.

[0103] In embodiments, an amino acid having a side chain comprising a guanidine replacement group or a protonated form thereof, can be adjacent to an amino acid having a side chain comprising an aryl or heteroaryl group. In embodiments, an amino acid having a side chain comprising a guanidine replacement group or a protonated form thereof, can be adjacent to at least one amino acid having a side chain comprising a guanidine or protonated form thereof. In embodiments, an amino acid having a side chain comprising a guanidine or protonated form thereof can be adjacent to an amino acid having a side chain comprising an aryl or heteroaryl group. In embodiments, two amino acids having a side chain comprising a guanidine replacement group or protonated forms thereof can be adjacent to each other. In embodiments, two amino acids having a side chain comprising a guanidine or protonated form thereof are adjacent to each other. In embodiments, a cCPP can comprise at least two contiguous amino acids having a side chain can comprise an aryl or heteroaryl group and at least two non-adjacent amino acids having a side chain comprising a guanidine replacement group or a protonated form thereof. In embodiments, a cCPP can comprise at least two contiguous amino acids having a side chain comprising an aryl or heteroaryl group and at least two non-adjacent amino acids having a side chain comprising

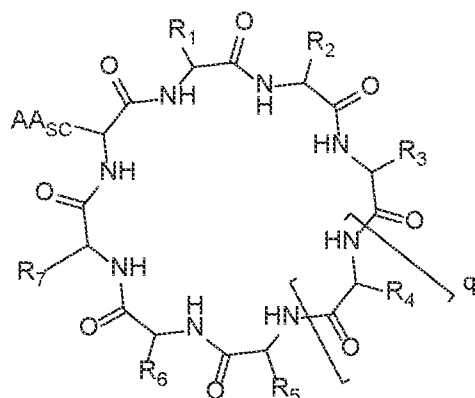


, or a protonated form thereof. In embodiments, the adjacent amino acids can have the same chirality. In embodiments, the adjacent amino acids can have the opposite chirality. Other combinations of amino acids can have any arrangement of D and L amino acids, e.g., any of the sequences described in the preceding paragraph.

[0104] In embodiments, at least two amino acids having a side chain comprising a guanidine replacement group or a protonated form thereof, are alternating with at least two amino acids having a side chain comprising a guanidine group or protonated form thereof.

[0105] In embodiments, the cCPP can comprise the structure of Formula (1):

Formula (1)



(1), or a protonated form thereof,

wherein:

R₁, R₂, R₃, R₄, R₅, R₆, and R₇ are independently H or an amino acid side chain;

5 AAsc is an amino acid side chain; and

q is 1, 2, 3, or 4.

[0106] The cCPP of Formula (1) can have any configuration and/or amino acid side chain, including, but not limited to those described in PCT Publication Nos: WO 2015/179691, filed May 21, 2015, entitled “CELL PENETRATING PEPTIDES AND METHODS OF MAKING AND
10 USING THEREOF”; WO 2021/127650, filed December 21, 2020, entitled “COMPOSITIONS FOR DELIVERY OF ANTISENSE COMPOUNDS”; WO 2022/213118, filed March 31, 2022, entitled “CYCLIC CELL PENETRATING PEPTIDES”; WO 2022/241408, filed May 9, 2022, entitled “COMPOSITIONS AND METHODS FOR MODULATING TISSUE DISTRIBUTION OF INTRACELLULAR THERAPEUTICS”, U.S. Patent Publication No. 2023/0312653, filed
15 March 30, 2023, entitled “CYCLIC CELL PENETRATING PEPTIDES”; and US Patent No. 11,225,506, filed April 20, 2020, entitled “CELL PENETRATING PEPTIDES AND METHODS OF MAKING AND USING THEREOF”, the disclosures of each of which are hereby incorporated by reference herein in their entireties.

[0107] In embodiments, the cCPP are of Formula (1) or a protonated form thereof, wherein:

20 R₁, R₂, and R₃ are each independently H or an aryl or heteroaryl side chain of an amino acid;

at least two of R₁, R₂, and R₃ is an aryl or heteroaryl side chain of an amino acid;

R₄, R₅, R₆, R₇ are independently H or an amino acid side chain;

AAsc is an amino acid side chain; and

q is 1, 2, 3, or 4.

[0108] In embodiments, the cCPP are of Formula (1) or a protonated form thereof, wherein:

R₁, R₂, and R₃ are each independently H or an aryl or heteroaryl side chain of an amino acid;

at least two of R₁, R₂, and R₃ is an aryl or heteroaryl side chain of an amino acid;

R₄, R₅, R₆, R₇ are independently H or an amino acid side chain;

5 at least two of R₄, R₅, R₆, R₇ are, independently, a side chain of arginine;

AA_{SC} is an amino acid side chain; and

q is 1, 2, 3, or 4.

[0109] In embodiments of Formula (1),

R₁, R₂, and R₃ are each independently H or a side chain comprising an aryl or heteroaryl group;

10 at least two of R₁, R₂, and R₃ are a side chain of phenylalanine;

R₄, R₅, R₆, and R₇ are independently H or an amino acid side chain;

AA_{SC} is an amino acid side chain; and

q is 1, 2, 3, or 4.

[0110] In embodiments of Formula (1),

15 one of R₁, R₂, and R₃ is H;

two of R₁, R₂, and R₃ are CH₂Ph; and

R₄, R₅, R₆, and R₇ are, independently, H or an amino acid side chain.

[0111] In embodiments of Formula (1),

R₁, R₂, and R₃ are -CH₂Ph;

20 R₄, R₅, R₆, and R₇ are, independently, H or an amino acid side chain.

[0112] In embodiments of Formula (1),

R₁, R₂, and R₃ are each independently a side chain comprising an aryl or heteroaryl group;

at least two of R₁, R₂, and R₃ are a side chain of phenylalanine;

R₄, R₅, R₆, and R₇ are, independently, H or an amino acid side chain; and

25 q is 1, 2, 3 or 4.

[0113] In embodiments of Formula (1),

R₁, R₂, and R₃ are each independently H or an amino acid side chain;

at least one of R₁, R₂, and R₃ is an aryl or heteroaryl side chain of an amino acid;

at least one of R₁, R₂, and R₃ are a side chain of arginine;

30 R₄, R₅, R₆, and R₇ are, independently, H or an amino acid side chain; and

q is 1, 2, 3 or 4.

[0114] In embodiments of Formula (1),

R_1 , R_2 , and R_3 are each independently H or an amino acid side chain;

at least two of R_1 , R_2 , and R_3 are a side chain of arginine;

R_4 , R_5 , R_6 , and R_7 are, independently, H or an amino acid side chain; and

5 q is 1, 2, 3 or 4.

[0001] In embodiments of Formula (I),

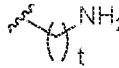
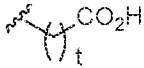
R_1 , R_2 , and R_3 are each independently a side chain comprising an aryl or heteroaryl group;

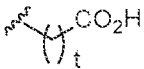
at least two of R_1 , R_2 , and R_3 are a side chain of naphthylalanine;

R_4 , R_5 , R_6 , and R_7 are, independently, H or an amino acid side chain; and

10 q is 1, 2, 3, or 4.

[0002] AA_{SC} can be a side chain or terminus of an amino acid residue on the cCPP. Non-limiting examples of AA_{SC} include aspartic acid, glutamic acid, glutamine, asparagine, or lysine, or a modified side chain of glutamine or asparagine (e.g., a reduced side chain having an amino group).

AA_{SC} can be an AA_{SC} as defined herein. In embodiments, AA_{SC} can be  or .

15 wherein t can be an integer from 0 to 5. AA_{SC} can be , wherein t can be 0 or an integer from 1 to 5. t can be 1 to 5. t is 2 or 3. t can be 2. t can be 3. In embodiments, AA_{SC} can be conjugated to a linker.

[0003] In embodiments the cCPP is of Formula (1), where at least one of R_4 , R_5 , R_6 , R_7 are independently an uncharged, non-aromatic side chain of an amino acid. In embodiments, at least
20 one of R_4 , R_5 , R_6 , and R_7 are independently H or a side chain of serine, histidine or citrulline. In embodiments, one of R_4 , R_5 , R_6 , and R_7 are independently H or a side chain of serine, histidine or citrulline. In embodiments, at least two of R_4 , R_5 , R_6 , and R_7 are independently H or a side chain of serine, histidine or citrulline. In embodiments, two of R_4 , R_5 , R_6 , and R_7 are independently H or a side chain of serine, histidine or citrulline.

25 [0004] In embodiments, compounds are provided that include a cCPP having from 6 to 10 amino acids, wherein at least two amino acids of the cCPP are charged amino acids, at least two amino acids of the cCPP are aromatic hydrophobic amino acids and at least two amino acids of the cCPP are uncharged, non-aromatic amino acids. In embodiments, at least two charged amino acids of the cCPP are arginine. In embodiments, two charged amino acids of the cCPP are arginine. In

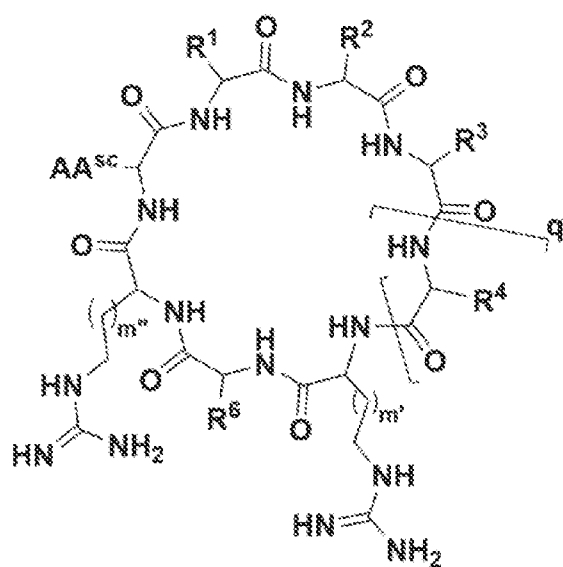
embodiments, at least two aromatic, hydrophobic amino acids of the cCPP are, independently, phenylalanine, 2-naphthylalanine, or a combination thereof. In embodiments, two or three aromatic, hydrophobic amino acids of the cCPP are, independently, phenylalanine, 2-naphthylalanine, or a combination thereof. In embodiments, at least two uncharged, non-aromatic amino acids of the cyclic peptide are citrulline, histidine, serine, glycine, or a combination thereof.

In embodiments, two amino acids of the cCPP are citrulline, histidine, serine, glycine or a combination thereof. In embodiments, the compound is a cCPP having from 6 to 10 amino acids wherein two amino acids of the cCPP are arginine, two or three amino acids are, independently, aromatic, hydrophobic amino acids selected from phenylalanine, 2-naphthylalanine, β -homophenylalanine, and two amino acids are independently, uncharged, non-aromatic amino acids selected from citrulline, serine, histidine, and glycine.

[0005] In embodiments the cCPP is of Formula (1), where at least two of R_4 , R_5 , R_6 , and R_7 are a positively charged side chain of an amino acid residue. In embodiments, two of R_4 , R_5 , R_6 , and R_7 are a positively charged side chain of an amino acid residue. In embodiments, at least three of R_4 , R_5 , R_6 , and R_7 are a positively charged side chain of an amino acid residue. In embodiments, three of R_4 , R_5 , R_6 , R_7 are a positively charged side chain of an amino acid residue. In embodiments, R_4 , R_5 , R_6 , and R_7 are a positively charged side chain of an amino acid residue.

[0006] The cCPP can comprise the structure of Formula (2):

Formula (2)



20

(2), or a protonated form thereof,

wherein:

R_1 , R_2 , and R_3 can each independently be H or an amino acid residue having a side chain comprising an aryl or heteroaryl group;

at least two of R_1 , R_2 , and R_3 are an aryl or heteroaryl side chain of an amino acid;

R_4 and R_6 are independently H or an amino acid side chain;

5 AA_{SC} is an amino acid side chain;

m' is an integer from 0 to 3;

m'' is an integer from 0 to 3; and

q is 1, 2, 3, or 4.

[0007] In embodiments, the cCPP are of Formula (1) or (2), where R_1 , R_2 , and R_3 can each
 10 independently be H, -alkylene-aryl, -alkylene-heteroaryl, or an amino acid side chain comprising a guanidine group, a guanidine replacement group, or a protonated form thereof. R_1 , R_2 , and R_3 can each independently be H, -C₁₋₃alkylene-aryl, -C₁₋₃alkylene-heteroaryl or an amino acid side chain comprising a guanidine group, or a protonated form thereof. In embodiments, the cCPP are of Formula (1) or (2), where R_1 , R_2 , and R_3 can each independently be H, -alkylene-aryl, or -
 15 alkylene-heteroaryl. R_1 , R_2 , and R_3 can each independently be H, -C₁₋₃alkylene-aryl, or -C₁₋₃alkylene-heteroaryl. R_1 , R_2 , and R_3 can each independently be H or -alkylene-aryl. R_1 , R_2 , and R_3 can each independently be H or -C₁₋₃alkylene-aryl. C₁₋₃alkylene can be methylene. Aryl can be a 6- to 14-membered aryl. Heteroaryl can be a 6- to 14-membered heteroaryl having one or more heteroatoms selected from N, O, and S. Aryl can be selected from phenyl, naphthyl, or anthracenyl.
 20 Aryl can be phenyl or naphthyl. Aryl can be phenyl. Heteroaryl can be pyridyl, quinolyl, and isoquinolyl. The amino acid side chain comprising a guanidine group can be arginine. R_1 , R_2 , and R_3 can each independently be H, -C₁₋₃alkylene-Ph or -C₁₋₃alkylene-naphthyl. R_1 , R_2 , and R_3 can each independently be H, -CH₂Ph, or -CH₂-naphthyl. R_1 , R_2 , and R_3 can each independently be H or -CH₂Ph. One of R_1 , R_2 , and R_3 can be arginine.

25 [0008] In embodiments, the cCPP are of Formula (1) or (2), where R_1 , R_2 , and R_3 can each independently be the side chain of tyrosine, phenylalanine, 1-naphthylalanine, 2-naphthylalanine, tryptophan, 3-benzothienylalanine, 4-phenylphenylalanine, 3,4-difluorophenylalanine, 4-trifluoromethylphenylalanine, 2,3,4,5,6-pentafluorophenylalanine, homophenylalanine, β -homophenylalanine, 4-tert-butyl-phenylalanine, 4-pyridinylalanine, 3-pyridinylalanine, 4-
 30 methylphenylalanine, 4-fluorophenylalanine, 4-chlorophenylalanine, 3-(9-anthryl)-alanine or

arginine. R₁, R₂, and R₃ can each independently be the side chain of tyrosine, phenylalanine, 1-naphthylalanine, 2-naphthylalanine, arginine or tryptophan.

[0009] In embodiments, the cCPP are of Formula (1) or (2), where R₁ can be the side chain of tyrosine. R₁ can be the side chain of phenylalanine. R₁ can be the side chain of 1-naphthylalanine.

5 R₁ can be the side chain of 2-naphthylalanine. R₁ can be the side chain of tryptophan. R₁ can be the side chain of 3-benzothienylalanine. R₁ can be the side chain of 4-phenylphenylalanine. R₁ can be the side chain of 3,4-difluorophenylalanine. R₁ can be the side chain of 4-trifluoromethylphenylalanine. R₁ can be the side chain of 2,3,4,5,6-pentafluorophenylalanine. R₁ can be the side chain of homophenylalanine. R₁ can be the side chain of β -homophenylalanine. R₁
10 can be the side chain of 4-tert-butyl-phenylalanine. R₁ can be the side chain of 4-pyridinylalanine. R₁ can be the side chain of 3-pyridinylalanine. R₁ can be the side chain of 4-methylphenylalanine. R₁ can be the side chain of 4-fluorophenylalanine. R₁ can be the side chain of 4-chlorophenylalanine. R₁ can be the side chain of 3-(9-anthryl)-alanine. R₁ can be H. R₁ can be a side chain of arginine.

15 [0010] In embodiments, the cCPP are of Formula (1) or (2), where R₂ can be the side chain of tyrosine. R₂ can be the side chain of phenylalanine. R₂ can be the side chain of 1-naphthylalanine. R₂ can be the side chain of 2-naphthylalanine. R₂ can be the side chain of tryptophan. R₂ can be the side chain of 3-benzothienylalanine. R₂ can be the side chain of 4-phenylphenylalanine. R₂ can be the side chain of 3,4-difluorophenylalanine. R₂ can be the side chain of 4-trifluoromethylphenylalanine. R₂ can be the side chain of 2,3,4,5,6-pentafluorophenylalanine. R₂
20 can be the side chain of homophenylalanine. R₂ can be the side chain of β -homophenylalanine. R₂ can be the side chain of 4-tert-butyl-phenylalanine. R₂ can be the side chain of 4-pyridinylalanine. R₂ can be the side chain of 3-pyridinylalanine. R₂ can be the side chain of 4-methylphenylalanine. R₂ can be the side chain of 4-fluorophenylalanine. R₂ can be the side chain of 4-chlorophenylalanine. R₂ can be the side chain of 3-(9-anthryl)-alanine. R₂ can be H. R₂ can be a side chain of arginine.

[0011] In embodiments, the cCPP are of Formula (1) or (2), where R₃ can be the side chain of tyrosine. R₃ can be the side chain of phenylalanine. R₃ can be the side chain of 1-naphthylalanine. R₃ can be the side chain of 2-naphthylalanine. R₃ can be the side chain of tryptophan. R₃ can be
30 the side chain of 3-benzothienylalanine. R₃ can be the side chain of 4-phenylphenylalanine. R₃ can be the side chain of 3,4-difluorophenylalanine. R₃ can be the side chain of 4-

trifluoromethylphenylalanine. R₃ can be the side chain of 2,3,4,5,6-pentafluorophenylalanine. R₃ can be the side chain of homophenylalanine. R₃ can be the side chain of β-homophenylalanine. R₃ can be the side chain of 4-tert-butyl-phenylalanine. R₃ can be the side chain of 4-pyridinylalanine. R₃ can be the side chain of 3-pyridinylalanine. R₃ can be the side chain of 4-methylphenylalanine.

5 R₃ can be the side chain of 4-fluorophenylalanine. R₃ can be the side chain of 4-chlorophenylalanine. R₃ can be the side chain of 3-(9-anthryl)-alanine. R₃ can be H. R₃ can be a side chain of arginine.

[0012] In embodiments, the cCPP are of Formula (1) or (2), where R₄ can be H, or a side chain of arginine, citrulline, serine, or histidine. R₄ can be H or a side chain of arginine. R₄ can be H. R₄ can be a side chain of arginine. R₄ can be a side chain of citrulline. R₄ can be a side chain of serine.

10 R₄ can be a side chain of histidine. R₄ can be a side chain of valine. R₄ can be a side chain of leucine.

[0013] In embodiments, the cCPP are of Formula (1), where R₅ can be H, or a side chain of arginine, citrulline, serine, or histidine. R₅ can be H or a side chain of arginine. R₅ can be H. R₅ can be a side chain of arginine. R₅ can be a side chain of citrulline. R₅ can be a side chain of serine.

15 R₅ can be a side chain of histidine.

[0014] In embodiments, the cCPP are of Formula (1) or (2), where R₆ can be H, or a side chain of arginine, citrulline, serine, or histidine. R₆ can be H or a side chain of arginine. R₆ can be H. R₆ can be a side chain of arginine. R₆ can be a side chain of citrulline. R₆ can be a side chain of serine.

20 R₆ can be a side chain of histidine. R₆ can be a side chain of valine. R₆ can be a side chain of leucine.

[0015] In embodiments, the cCPP are of Formula (2), where R₇ can be H, or a side chain of arginine, citrulline, serine, or histidine. R₇ can be H or a side chain of arginine. R₇ can be H. R₇ can be a side chain of arginine. R₇ can be a side chain of citrulline. R₇ can be a side chain of serine.

25 R₇ can be a side chain of histidine.

[0016] In embodiments, the cCPP are of Formula (1) or (2), where one, two, or three of R₁, R₂, R₃, R₄, R₅, R₆, and R₇ can be H. At least one of R₁, R₂, R₃, R₄, R₅, R₆, and R₇ can be H. One of R₁, R₂, R₃, R₄, R₅, R₆, and R₇ can be H. Two of R₁, R₂, R₃, R₄, R₅, R₆, and R₇ can be H. Three of R₁, R₂, R₃, R₅, R₆, and R₇ can be H. One of R₁, R₂, and R₃ can be H. At least one of R₄, R₅, R₆, and R₇ can be H. One of R₄, R₅, R₆, and R₇ can be H. Two of R₄, R₅, R₆, and R₇ can be H. Three of R₄, R₅, R₆, and R₇ can be H. Four of R₄, R₅, R₆, and R₇ can be H.

30

[0017] In embodiments, the cCPP are of Formula (1) or (2), where at least one of R_4 , R_5 , R_6 , and R_7 can be H or a side chain of arginine, citrulline, serine, or histidine. At least one of R_4 , R_5 , R_6 , and R_7 is H. At least one of R_4 , R_5 , R_6 , and R_7 can be side chain of arginine. At least one of R_4 , R_5 , R_6 , and R_7 can be side chain of citrulline. At least one of R_4 , R_5 , R_6 , and R_7 can be side chain of serine. At least one of R_4 , R_5 , R_6 , and R_7 can be side chain of histidine. One of R_4 , R_5 , R_6 , and R_7 can be H or a side chain of arginine, citrulline, serine, or histidine. One of R_4 , R_5 , R_6 , and R_7 is H. One of R_4 , R_5 , R_6 , and R_7 can be side chain of arginine. One of R_4 , R_5 , R_6 , and R_7 can be side chain of citrulline. One of R_4 , R_5 , R_6 , and R_7 can be side chain of serine. One of R_4 , R_5 , R_6 , and R_7 can be side chain of histidine.

[0018] In embodiments, the cCPP are of Formula (1) or (2), where two of R_4 , R_5 , R_6 , and R_7 can be H or a side chain of arginine, citrulline, serine, or histidine. Two of R_4 , R_5 , R_6 , and R_7 can be H. Two of R_4 , R_5 , R_6 , and R_7 can be side chain of arginine. Two of R_4 , R_5 , R_6 , and R_7 can be side chain of citrulline. Two of R_4 , R_5 , R_6 , and R_7 can be side chain of serine. Two of R_4 , R_5 , R_6 , and R_7 can be side chain of histidine.

[0019] In embodiments, the cCPP are of Formula (1) or (2), where three of R_4 , R_5 , R_6 , and R_7 can be H or a side chain arginine, citrulline, serine or histidine. Three of R_4 , R_5 , R_6 , and R_7 can be H. Three of R_4 , R_5 , R_6 , and R_7 can be side chain of arginine. Three of R_4 , R_5 , R_6 , and R_7 can be side chain of citrulline. Three of R_4 , R_5 , R_6 , and R_7 can be side chain of serine. Three of R_4 , R_5 , R_6 , and R_7 can be side chain of histidine.

[0020] In embodiments, the cCPP are of Formula (1) or (2), where AA_{SC} can be a side chain of a residue of asparagine, glutamine, or homoglutamine. AA_{SC} can be a side chain of a residue of

glutamine. In embodiments, AA_{SC} can be $\text{---}(\text{---})_t\text{NH}_2$ or $\text{---}(\text{---})_t\text{CO}_2\text{H}$, wherein t can be an integer

from 0 to 5. AA_{SC} can be $\text{---}(\text{---})_t\text{CO}_2\text{H}$, wherein t can be 0 or an integer from 1 to 5. t can be 1 to 5. t is 2 or 3. t can be 2. t can be 3.

[0021] In embodiments, the cCPP are of Formula (1) or (2), where q can be 1, 2, or 3. q can be 1 or 2. q can be 1. q can be 2. q can be 3. q can be 4.

[0022] In embodiments, the cCPP are of Formulae (1) or (2), where m' can be 1 to 3. m' can be 1 or 2. m' can be 0. m' can be 1. m' can be 2. m' can be 3.

[0023] In embodiments, the cCPP are of Formulae (1) or (2), where m'' can be 1 to 3. m'' can be 1 or 2. m'' can be 0. m'' can be 1. m'' can be 2. m'' can be 3.

[0024] In embodiments, the cCPP comprises the structure of Formula (2) or a protonated form thereof, wherein:

- 5 one of R_1 , R_2 , and R_3 is H;
- two of R_1 , R_2 , and R_3 are $-\text{CH}_2\text{Ph}$;
- R_4 and R_6 are independently H or an amino acid side chain;
- AA_{sc} is an amino acid side chain;
- q is 1, 2, 3 or 4;

- 10 m' is an integer from 0 to 3; and
- m'' is an integer from 0 to 3.

[0025] In embodiments, the cCPP comprises the structure of Formula (2) or a protonated form thereof, wherein:

- R_1 , R_2 , and R_3 are $-\text{CH}_2\text{Ph}$;
- 15 R_4 and R_6 are independently H or an amino acid side chain;
- AA_{sc} is an amino acid side chain;
- q is 1, 2, 3 or 4;
- m' is an integer from 0 to 3; and

- [0026] m'' is an integer from 0 to 3. In embodiments, the cCPP comprises the structure of Formula
- 20 (2) or a protonated form thereof, wherein:

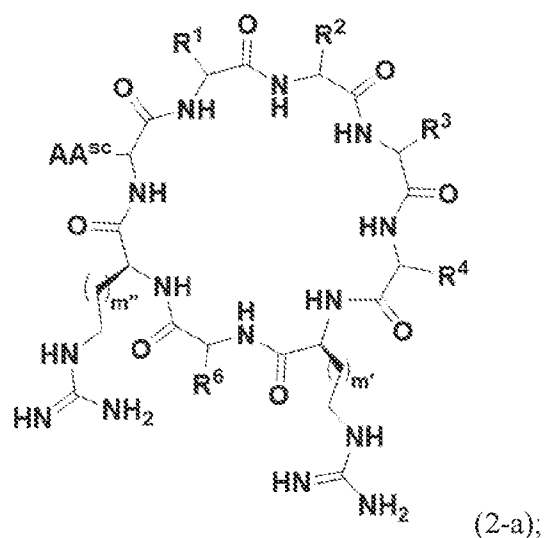
- R_1 , R_2 , and R_3 are each independently a side chain of an amino acid comprising an aryl or heteroaryl group;
- at least one of R_1 , R_2 , and R_3 are a side chain of naphthylalanine;
- AA_{sc} is an amino acid side chain;
- 25 R_4 and R_6 are independently H or an amino acid side chain of arginine, serine, histidine or citrulline;
- q is 1, 2, 3 or 4;
- m' is an integer from 0 to 3; and

- [0027] m'' is an integer from 0 to 3. In embodiments, the cCPP comprises the structure of Formula
- 30 (2) or a protonated form thereof, and R_4 and R_6 are independently H, or a side chain of arginine, histidine, serine or citrulline. In embodiments, R_4 and R_6 are independently H, or a side chain

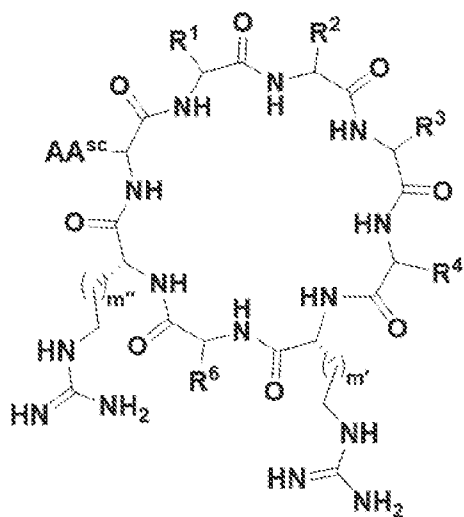
of arginine, histidine, or serine. In embodiments, R_4 is H. In embodiments, R_4 is a side chain of arginine. In embodiments, R_4 is a side chain of histidine. In embodiments, R_4 is a side chain of serine. In embodiments, R_4 is a side chain of citrulline. In embodiments, R_6 is H. In embodiments, R_6 is a side chain of arginine. In embodiments, R_6 is a side chain of histidine. In embodiments, R_6 is a side chain of serine. In embodiments, R_6 is a side chain of citrulline.

[0028] The cCPP of Formula (2) can comprise the structure of Formula (2-a) or Formula (II-b):

Formula (2-a)



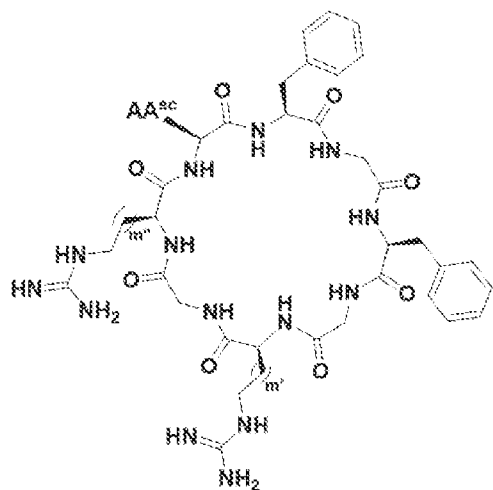
Formula (2-b)



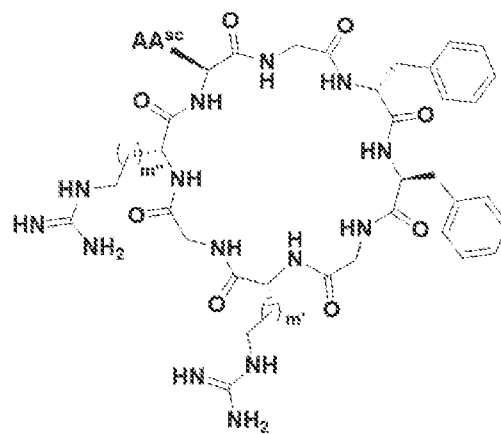
wherein AA^{sc} , R_1 , R_2 , R_3 , R_4 , R_6 , m' , and m'' are as defined herein relative to Formula (2).

[0029] The cCPP of Formula (2) can comprise the structures of Formulae (2-a1), (2-b1), (2-a2), (2-b2), (2-a3), or (2-b3):

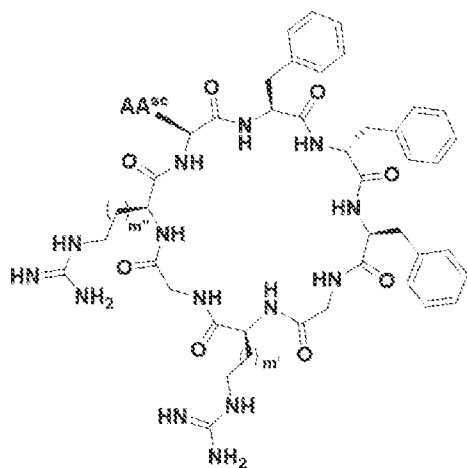
Formulae (2-a1), (2-b1), (2-a2), (2-b2), (2-a3), or (2-b3):



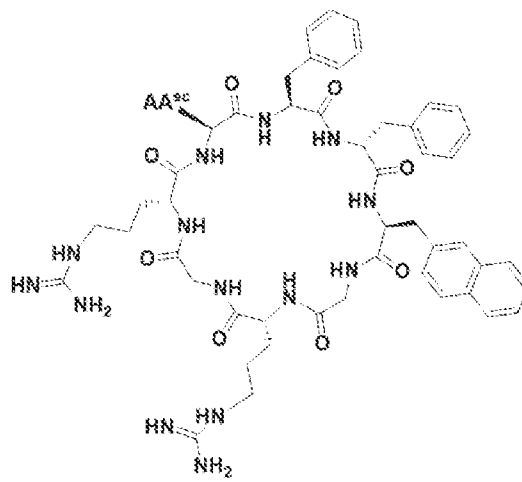
(2-a1);



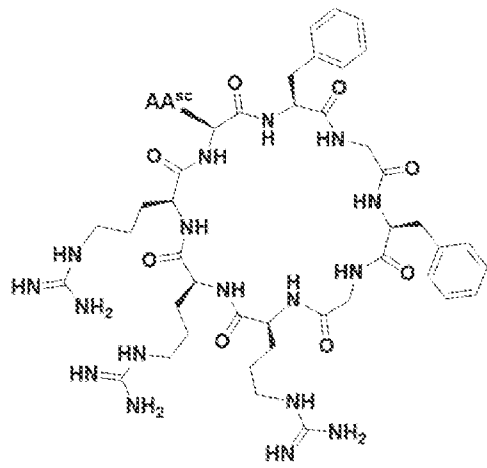
(2-b1);



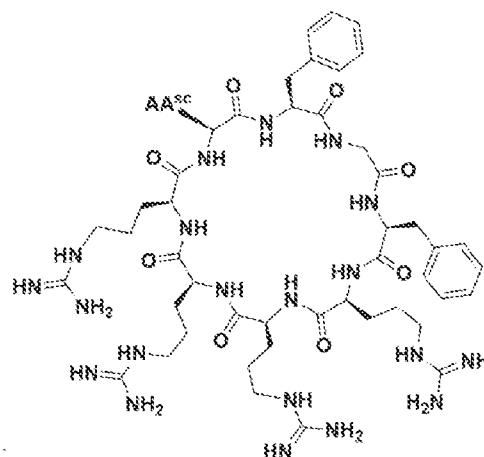
(2-a2);



(2-b2);



(2-a3);



(2-b3); or

protonated form thereof, wherein AA_{SC}, m', and m'' are as defined herein relative to Formula (2).

[0030] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: FGFGRRGR; GfFGrrGr; FffGRRGR; FGFGRRRR; or FGFRRRR.

[0031] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: FGFGHGH or FGFSHSH.

[0032] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: FfFSRSR or FGFSRSR.

[0033] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: Ff-Nal-RrRr; Ff-Nal-GrGr; Ff-Nal-GRGR; Ff-Nal-HrHr; or Ff-Nal-SrSr.

[0034] In embodiments, the cCPP of Formula (1) or (2) can comprise one of the following sequences: Ff-Nal-Cit-r-Cit-r; Ff-Nal-Rr-Cit-r; Ff-Nal-Cit-rRr; or Ff-Nal-R-cit-R-cit.

[0035] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: FGFGRRGRQ; GfFGrrGrQ; FffGRRGRQ; FGFGRRRRQ; or FGFRRRRQ.

[0036] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: FGFGHGHQ or FGFSHSHQ.

[0037] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: FfFSRSRQ or FGRSRSRQ.

[0038] In embodiments, the cCPP of Formula (1) or (2) can include those having one of the following sequences: Ff-Nal-RrRrQ; Ff-Nal-GrGrQ; Ff-Nal-GRGRQ; Ff-Nal-HrHrQ; or Ff-Nal-SrSrQ.

[0039] In embodiments, the cCPP of Formula (1) or (2) can comprise one of the following sequences: Ff-Nal-Cit-r-Cit-rQ; Ff-Nal-Rr-Cit-rQ; Ff-Nal-Cit-rRrQ; or Ff-Nal-R-cit-R-cit-Q.

cCPP sequence		
F-Nal-RRRQ	FRRRR-Nal-Q	F-Nal-RRRRQK
F-Nal-RRRC	rRFR-Nal-RQ	F-Nal-RRRRQC
F-Nal-RRRU	RR-Nal-FRRQ	f-Nal-RrRrRQ
RRR-Nal-FQ	CRRRRFWQ	F-Nal-RRRRRQ
RRRR-Nal-F	Ff-Nal-RrRrQ	RRRR-Nal-FDΩC
F-Nal-RRRR	FF-Nal-RRRRQ	F-Nal-RRR
F-nal-rRrRq	RFRFR-Nal-RQ	FWRRR
F-nal-rRrRQ	URRRRFWQ	RRR-Nal-F
F-Nal-RRRRQ		RRRWF
f-Nal-RrRrQ		

RRFR-Nal-RQ		
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where Nal = L-naphthylalanine; nal = D-naphthylalanine; Ω = L-norleucine.

[0040] In embodiments, the cCPP of Formula (1) or (2) can be selected from Ff-Nal-GrGrQ; FfFGRGRQ; FGFGGRGRQ; GfFGrGrQ; FfFGRGRQ; FGFGRRRQ and FGFRRRRQ.

[0041] In embodiments, the cCPP of Formula (1) or (2) can be FAFARARQ.

5 [0042] In embodiments, the cCPP of Formula (1) or (2) can be Ff-Nal-GrGrQ.

[0043] In embodiments, the cCPP of Formula (1) or (2) can be selected from FfFGRGRQ, FfFRRRRQ, FfFRrRrQ, fffrrrrQ or FfFRrRrQ.

[0044] In embodiments, the cCPP of Formula (1) or (2) can be FG-Nal-GRGRQ.

10 [0045] In embodiments, the cCPP of Formula (1) or (2) can be selected from FGFGGRGRQ, fGfGrGrQ, fGfGrGrQ, FGFGGRGRQ, FGFGGRQ, fGfrrrrQ, FGFRRRRQ, or FGFRRRRQ.

[0046] In embodiments, the cCPP of Formula (I) or (II) can be selected from FFFRRRRQ, FFFGRRRQ, FFFRGRQ, FFFRRRGQ, GFFRRRRQ, or FFGRRRRQ.

[0047] In embodiments, the cCPP of Formula (I) or (II) can be FFFRRRRQ.

[0048] In embodiments, the cCPP of Formula (I) or (II) can be FFFGRRRQ.

15 [0049] In embodiments, the cCPP of Formula (I) or (II) can be FFFRGRQ.

[0050] In embodiments, the cCPP of Formula (I) or (II) can be FFFRRGRQ.

[0051] In embodiments, the cCPP of Formula (I) or (II) can be FFFRRRGQ.

[0052] In embodiments, the cCPP of Formula (I) or (II) can be GFFRRRRQ.

[0053] In embodiments, the cCPP of Formula (I) or (II) can be FFGRRRRQ.

20 [0054] In embodiments, the cCPP of Formula (I) or (II) can be selected from Nal-G-Nal-GRGRQ; FGFKRKRQ; FGFRRRRQ; FfGFRRRRQ; or FGFRRGRRQ.

[0055] In embodiments, the cCPP of Formula (I) or (II) can be Nal-G-Nal-GRGRQ.

[0056] In embodiments, the cCPP of Formula (I) or (II) can be FGFKRKRQ.

[0057] In embodiments, the cCPP of Formula (I) or (II) can be FGFRRRRQ.

25 [0058] In embodiments, the cCPP of Formula (I) or (II) can be FfGFRRRRQ.

[0059] In embodiments, the cCPP of Formula (I) or (II) can be FGFRRGRRQ.

[0060] In embodiments, the cCPP of Formula (I) or (II) can be selected from RGRGRGRQ; FFFGRGRQ; FGFRRRRQ; FGFRGRGQ; FRGRGRGQ; FRFGRGRQ; FRFRFRFQ; FGFLRLRQ; or FGFVRVRQ.

30 [0061] In embodiments, the cCPP of Formula (I) or (II) can be RGRGRGRQ.

[0062] In embodiments, the cCPP of Formula (I) or (II) can be FFFGRGRQ.

[0063] In embodiments, the cCPP of Formula (I) or (II) can be FGFRRRRQ.

[0064] In embodiments, the cCPP of Formula (I) or (II) can be FGFRGRGQ.

[0065] In embodiments, the cCPP of Formula (I) or (II) can be FRGRGRGQ.

5 [0066] In embodiments, the cCPP of Formula (I) or (II) can be FRFRGRGRQ.

[0067] In embodiments, the cCPP of Formula (I) or (II) can be FRFRFRFQ.

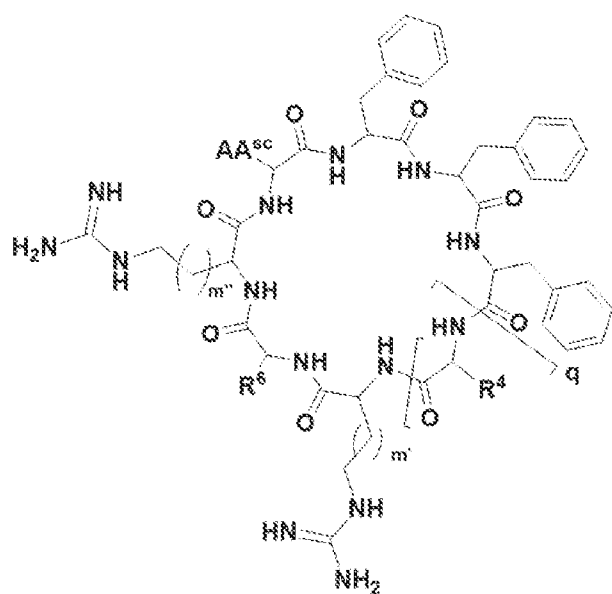
[0068] In embodiments, the cCPP of Formula (I) or (II) can be FGFLRLRQ.

[0069] In embodiments, the cCPP of Formula (I) or (II) can be FGFVRVRQ. In embodiments, the cCPP of Formula (1) or (2) can be FGWRGRQ.

10 [0070] In embodiments, the cCPP of Formula (1) or (2) can be fFRGRQ.

[0071] The cCPP of Formula (2) can have the structure of Formula (II-c):

Formula (2-c):

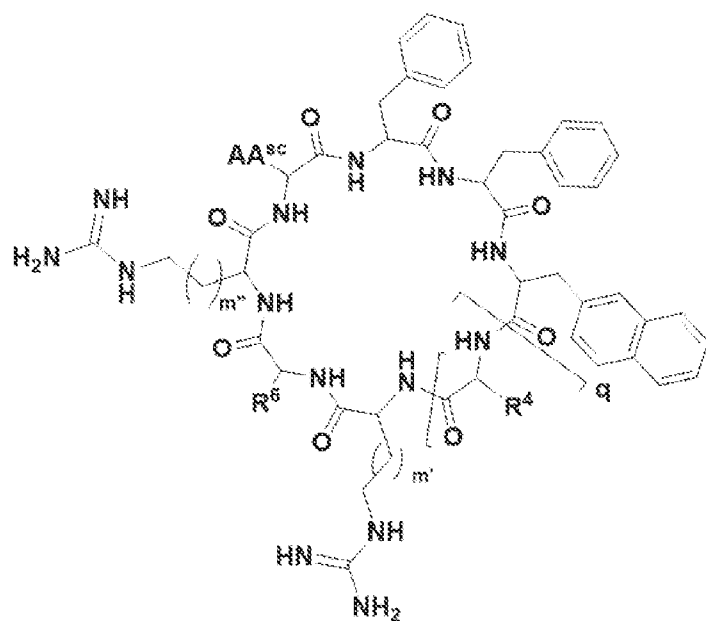


(2-c), or a protonated form thereof,

wherein R_4 , R_6 , q , m' , m'' , and AA^{sc} are as defined herein.

15 [0072] The cCPP of Formula (2) can have the structure of Formula (2-d):

Formula (2-d)

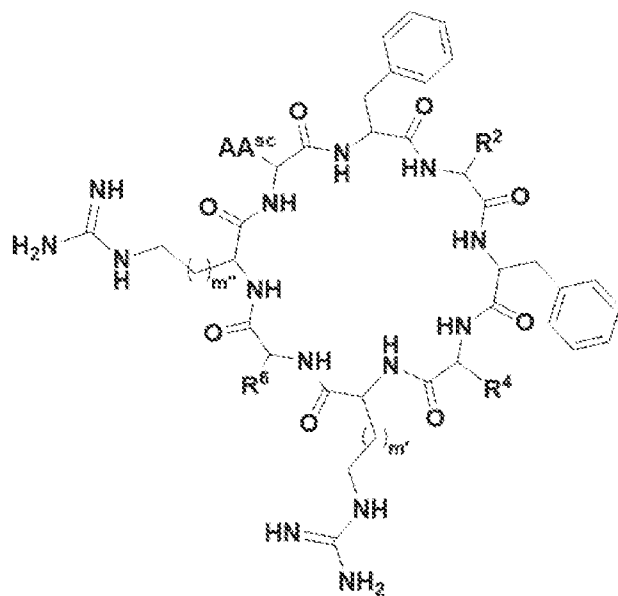


(2-d), or a protonated form thereof,

wherein R_4 , R_6 , q , m' , m'' , and AA^{sc} are as defined herein.

[0073] The cCPP of Formula (2) can have the structure of Formula (2-e):

5 Formula (2-e):

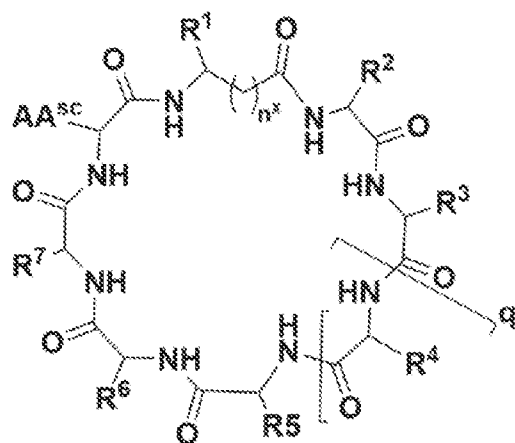


(2-e), or a protonated form thereof,

wherein: R_2 , R_4 , R_6 , m' , m'' , and AA^{sc} are as defined herein. In embodiments, R_2 is H.

[0074] In embodiments, the cCPP can be of the Formula (3):

Formula (3):



(3), or a protonated form thereof,

wherein:

R₁, R₂, and R₃ can each independently be H or an amino acid residue having a side chain

5 comprising an aryl or heteroaryl group;

at least two of R₁, R₂, and R₃ is an aryl or heteroaryl side chain of an amino acid;

R₄, R₅, R₆, and R₇ are independently H or an amino acid side chain;

at least two of R₄, R₅, R₆, and R₇ are independently a side chain of arginine;

AA_{sc} is an amino acid side chain;

10 n^x is 0 or 1; and

q is 1, 2, 3, or 4.

[0075] In embodiments, the cCPP is of Formula (3) where at least one of R₁, R₂, or R₃ is H. In
embodiments, the amino acid residue having a side chain comprising an aryl or heteroaryl group
is phenylalanine, homophenylalanine, or 2-naphthylalanine. In embodiments, the cCPP is of
15 Formula (3) where at least two of R₄, R₅, R₆, or R₇ are each independently an amino acid residue
having a side chain comprising a charged group. In embodiments, the amino acid residue having
a side chain comprising a charged group is arginine. In embodiments, the cCPP is of Formula (3)
where q is 1.

[0076] In embodiments, the cCPP is of Formula (3) where n^x is 1, where R₅ and R₇ are a side
20 chain of arginine. In embodiments, the cCPP is of Formula (3) where n^x is 1, wherein the aryl or
heteroaryl group is phenylalanine, beta-homophenylalanine, or 2-naphthylalanine, and where at
least two of R₄, R₅, R₆ and R₇ are the side chain of arginine. In embodiments, the cCPP is of
Formula (3) where n^x is 1, where R₄, R₅, R₆ and R₇ are the side chain of arginine. In embodiments,

the cCPP is of Formula (3) where n^X is 1, where R_5 and R_7 are the side chain of arginine, and R_4 and R_6 are H.

[0077] In embodiments, the cCPP is of Formula (3) where at least one of R_4 , R_5 , R_6 , or R_7 is the amino acid side chain of serine or histidine. In embodiments the cCPP is of Formula (3) where at least two of R_4 , R_5 , R_6 , or R_7 are, independently, the amino acid side chain of serine, citrulline, or histidine. In embodiments the cCPP is of Formula (3) where at least three of R_4 , R_5 , R_6 , or R_7 are, independently, the amino acid side chain of serine or histidine. In embodiments the cCPP is of Formula (3) where at least four of R_4 , R_5 , R_6 , or R_7 are, independently, the amino acid side chain of serine or histidine.

[0078] In embodiments of the cCPP of Formula (3):

at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, or 2-naphthylalanine;

at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of arginine;

at least two of R_4 , R_5 , R_6 , or R_7 are independently H, or a side chain of arginine, serine, citrulline, or histidine;

AA_{SC} is an amino acid side chain;

n^X is 0 or 1; and

q is 1.

[0079] In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 , R_6 , or R_7 are a side chain of serine. In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 , R_6 , or R_7 are a side chain of histidine. In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 , R_6 , or R_7 are a side chain of citrulline. In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 , R_6 , or R_7 are independently, H.

[0080] In embodiments, the cCPP is of Formula (3) wherein:

at least two of R_1 , R_2 , and R_3 are a side chain of phenylalanine or naphthylalanine;

at least two of R_4 , R_5 , R_6 , or R_7 are a side chain of arginine;

at least two of R_4 , R_5 , R_6 , or R_7 are independently H, or a side chain of an uncharged non-aryl amino acid selected from histidine, threonine, serine, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, citrulline, and 3-(4-thienyl)-alanine;

AA_{SC} is an amino acid side chain;

n^X is 0 or 1; and

q is 1.

[0081] In embodiments, the cCPP is of Formula (3) wherein:

at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine or naphthylalanine;

5 at least two of R_4 , R_5 , R_6 , or R_7 are a side chain of arginine;

at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of serine, citrulline or histidine;

AA_{SC} is an amino acid side chain;

n^X is 0 or 1; and

q is 1.

10 [0082] In embodiments, the cCPP is of Formula (3), wherein at least one of R_1 , R_2 , or R_3 is H. In embodiments, the cCPP is of Formula (3), wherein at least one of R_1 , R_2 , or R_3 is a side chain of phenylalanine. In embodiments, the cCPP is of Formula (3), wherein at least two of R_1 , R_2 , or R_3 are a side chain of phenylalanine. In embodiments, the cCPP is of Formula (3), wherein at least one of R_1 , R_2 , or R_3 is a side chain of 2-naphthylalanine.

15 [0083] In embodiments, the cCPP is of Formula (3), wherein at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of serine, citrulline or histidine.

[0084] In embodiments, the cCPP is of Formula (3), wherein at least one of R_4 , R_5 , R_6 , or R_7 are independently an uncharged, non-aryl side chain of an amino acid. In embodiments, at least two of R_4 , R_5 , R_6 , or R_7 are independently side chains of an uncharged non-aryl amino acid (e.g.,
20 histidine, citrulline, threonine, serine, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine). In embodiments, the cCPP is of Formula (3), wherein at least two of R_4 , R_5 , R_6 , or R_7 are independently side chains of an uncharged non-aryl amino acid selected from histidine, citrulline, threonine, serine, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine,
25 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine. In embodiments, the cCPP is of Formula (3), wherein at least two of R_4 , R_5 , R_6 , or R_7 are independently side chains of an uncharged non-aryl amino acid selected from histidine, citrulline, and serine.

[0085] In embodiments, the cCPP is of Formula (3), wherein at least one of R_4 , R_5 , R_6 , or R_7 is, independently, H. In embodiments, the cCPP is of Formula (3), wherein two of R_4 , R_5 , R_6 , or
30 R_7 are, independently, H.

[0086] In embodiments, the cCPP can be of Formula (3), wherein:

R_1 , R_2 , and R_3 can each independently be H or an amino acid residue having a side chain comprising an aryl or heteroaryl group;

at least two of R_1 , R_2 , and R_3 are phenylalanine;

R_4 , R_5 , R_6 , R_7 are independently H or an amino acid side chain;

5 two of R_4 , R_5 , R_6 , R_7 are independently a side chain of arginine;

AA_{SC} is an amino acid side chain;

n^X is 0 or 1; and

q is 1, 2, 3, or 4.

[0087] In embodiments, the cCPP is of Formula (3) where at least one of R_1 , R_2 , or R_3 are, independently, H. In embodiments, the cCPP is of Formula (3) where at least one of R_1 , R_2 , and R_3 are, independently, an amino acid residue having a side chain comprising an aryl or heteroaryl group. In embodiments, the amino acid residue having a side chain comprising an aryl or heteroaryl group is phenylalanine, β -homophenylalanine, or 2-naphthylalanine. In embodiments, the cCPP is of Formula (3) where at least two of R_4 , R_6 are each independently an amino acid residue having a side chain comprising a charged group. In embodiments, the amino acid residue having a side chain comprising a charged group is arginine. In embodiments, the cCPP is of Formula (3) where q is 1.

[0088] In embodiments, the cCPP is of Formula (3) where n^X is 1 and where R_5 and R_7 are a side chain of arginine. In embodiments, the cCPP is of Formula (3) where n^X is 1, wherein the aryl or heteroaryl group is phenylalanine, β -homophenylalanine, or 2-naphthylalanine, and where at least two of R_4 , R_5 , R_6 and R_7 are the side chain of arginine. In embodiments, the cCPP is of Formula (3) where n^X is 1, where R_4 , R_5 , R_6 and R_7 are the side chain of arginine. In embodiments, the cCPP is of Formula (3) where n^X is 1, where R_5 and R_7 are the side chain of arginine, and R_4 and R_6 are H. In embodiments, the cCPP is of Formula (3) where at least one of R_4 , R_5 (if present), R_6 , or R_7 (if present) are H or the amino acid side chain of serine, citrulline, or histidine. In embodiments the cCPP is of Formula (3) where at least two of R_4 , R_5 (if present), R_6 , or R_7 (if present) are, independently, H or the amino acid side chain of serine or histidine. In embodiments the cCPP is of Formula (3) where at least three of R_4 , R_5 , R_6 , or R_7 are, independently, H or the amino acid side chain of serine, citrulline, or histidine. In embodiments the cCPP is of Formula (3) where at least four of R_4 , R_5 , R_6 , or R_7 are, independently, H or the amino acid side chain of serine, citrulline, or histidine.

[0089] In embodiments of the cCPP of Formula (3): at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, b-homophenylalanine, or 2-naphthylalanine; at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of arginine; at least two of R_4 , R_5 , R_6 , or R_7 are independently H or a side chain of arginine, serine, citrulline, or histidine; AA_{SC} is an amino acid side chain; n^X is 0 or 1; and q is 1. It is understood that n^X is 1 when R_1 is a side chain of b-homophenylalanine.

[0090] In embodiments of the cCPP of Formula (3): at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, or 2-naphthylalanine; R_4 and R_6 are each independently H or a side chain of arginine, serine, citrulline or histidine; R_5 and R_7 are a side chain of arginine; AA_{SC} is an amino acid side chain; n^X is 0 or 1; and q is 1.

[0091] In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 (if present), R_6 , or R_7 (if present) are independently a side chain of serine. In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 (if present), R_6 , or R_7 (if present) are independently a side chain of histidine. In embodiments, the cCPP is of Formula (3), where two of R_4 , R_5 (if present), R_6 , or R_7 (if present) are independently, H.

[0092] In embodiments, the cCPP is of Formula (3) wherein: at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, beta-homophenylalanine, or naphthylalanine; at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of arginine; at least two of R_4 , R_5 , R_6 , or R_7 are independently H or a side chain of an uncharged non-aryl amino acid selected from histidine, threonine, serine, citrulline, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine; AA_{SC} is an amino acid side chain; n^X is 0 or 1; and q is 1. It is understood that n^X is 1 when R_1 is a side chain of b-homophenylalanine.

[0093] In embodiments, the cCPP is of Formula (3) wherein: at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, 2-naphthylalanine, or b-homophenylalanine; R_4 and R_6 are independently H or a side chain of an uncharged non-aryl amino acid selected from histidine, threonine, serine, citrulline, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine; R_5 and R_7 are a side chain of arginine; AA_{SC} is an amino acid side chain; n^X is 0 or 1; and q is 1. It is understood that n^X is 1 when R_1 is a side chain of b-homophenylalanine.

[0094] In embodiments, the cCPP is of Formula (3) wherein: at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, b-homophenylalanine, or 2-naphthylalanine; at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of arginine; at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of serine, citrulline or histidine; AA_{SC} is an amino acid side chain; n^X is 0 or 1; and q is 1. It is understood that n^X is 1 when R_1 is a side chain of b-homophenylalanine.

[0095] In embodiments, the cCPP is of Formula (3) wherein: at least two of R_1 , R_2 , and R_3 are independently a side chain of phenylalanine, beta-homophenylalanine, or naphthylalanine; R_4 and R_6 are independently a side chain of serine, citrulline or histidine; R_5 and R_7 are a side chain of arginine; AA_{SC} is an amino acid side chain; n^X is 0 or 1; and q is 1. It is understood that n^X is 1 when R_1 is a side chain of b-homophenylalanine.

[0096] In embodiments, the cCPP is of Formula (3), wherein at least one of R_1 , R_2 , and R_3 is H. In embodiments, the cCPP is of Formula (3), wherein at least one of R_1 , R_2 , and R_3 is a side chain of phenylalanine. In embodiments, the CPP is of Formula (3), wherein at least two of R_1 , R_2 , and R_3 are a side chain of phenylalanine. In embodiments, the CPP is of Formula (3), wherein two of R_1 , R_2 , and R_3 are a side chain of phenylalanine. In embodiments, the CPP is of Formula (3), wherein three of R_1 , R_2 , and R_3 are a side chain of phenylalanine. In embodiments, the cCPP is of the general Formula (3), wherein one of R_1 , R_2 , and R_3 is a side chain of naphthylalanine.

[0097] In embodiments, the cCPP is of Formula (3), wherein at least two of R_4 , R_5 , R_6 , or R_7 are independently a side chain of serine or histidine. In embodiments, the cCPP is of the general Formula (3), wherein R_4 and R_6 are independently H, or a side chain of serine, citrulline or histidine. In embodiments, the cCPP is of Formula (3), wherein R_4 and R_6 are the side chain of serine. In embodiments, the cCPP is of Formula (3), wherein R_4 and R_6 are the side chain of histidine. In embodiments, the cCPP is of Formula (3), wherein R_4 and R_6 are the side chain of citrulline. In embodiments, the cCPP is of Formula (3), wherein R_4 and R_6 are H. In embodiments, the cCPP is of Formula (3), wherein R_5 and R_7 are a side chain of arginine.

[0098] In embodiments, the cCPP is of Formula (3), wherein at least one of R_4 , R_5 (if present), R_6 , R_7 (if present) are independently an uncharged, non-aryl side chain of an amino acid. In embodiments, the cCPP is of Formula (3), wherein at least two of R_4 , R_5 (if present), R_6 , R_7 (if present) are independently side chains of an uncharged non-aryl amino acid (e.g., histidine, threonine, serine, citrulline, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine). In

embodiments, the cCPP is of Formula (3), wherein at least two of R₄, R₅ (if present), R₆, R₇ (if present) are independently side chains of an uncharged non-aryl amino acid selected from histidine, threonine, serine, leucine, isoleucine, valine, neopentylglycine, alanine, homoalanine, homoserine, 3-(4-thiazolyl)-alanine, 3-(4-furanyl)-alanine, and 3-(4-thienyl)-alanine. In
5 embodiments, the CPP is of Formula (3), wherein at least two of R₄, R₅ (if present), R₆, R₇ (if present) are independently side chains of an uncharged non-aryl amino acid selected from histidine and serine. In embodiments, the cCPP is of Formula (3), wherein R₅ and R₇ are a side chain of arginine.

[0099] In embodiments, the cCPP is of Formula (3), wherein at least one of R₄, R₅, R₆, or R₇ is,
10 independently, H. In embodiments, the cCPP is of Formula (3), wherein two of R₄, R₅, R₆, or R₇ are, independently, H. In embodiments, the cCPP is of Formula (3), wherein at least one of R₄ or R₆ is, independently, H. In embodiments, the cCPP is of Formula (3), wherein R₄ and R₆ are H. In embodiments, the cCPP is of Formula (3), wherein R₅ and R₇ are a side chain of arginine.

[0100] In embodiments, compounds are provided that include a cCPP having from 6 to 10 amino
15 acids, wherein at least two amino acids of the cCPP are charged amino acids, at least two amino acids of the cCPP are aryl or heteroaryl hydrophobic amino acids and at least two amino acids of the cCPP are uncharged, non-aryl amino acids. In embodiments, at least two charged amino acids of the cCPP are arginine. In embodiments, at least two aryl or heteroaryl, hydrophobic amino acids of the cCPP are phenylalanine, naphthylalanine (3-naphth-2-yl-alanine), b-homophenylalanine, or
20 a combination thereof. In embodiments, at least two uncharged, non-aryl amino acids of the cCPP are glycine. In embodiments, two of the uncharged amino acids are serine, citrulline, histidine, or a combination thereof.

[0101] In embodiments, the cCPP of Formula (3) can include those having one of the following
25 sequences: hFf-Nal-GrGr; bhF-F-Nal-SRSR; bhF-F-Nal-GRGR; bhF-F-Nal-HRHR; bhF-f-Nal-GrGr; bhF-f-Nal-SRSR; bhF-f-Nal-SrSr or bhFf-Nal-HrHr (where bhF – b-homophenylalanine; hFf – homophenylalanine).

[0102] In embodiments, the cCPP of Formula (3) can include those having one of the following
sequences: hFf-Nal-GrGrQ; bhF-F-Nal-SRSRQ; bhF-F-Nal-GRGRQ; bhF-F-Nal-HRHRQ; bhF-f-Nal-GrGrQ; bhF-f-Nal-SRSRQ; bhF-f-Nal-SrSrQ or bhFf-Nal-HrHrQ (where bhF – b-
30 homophenylalanine; hFf – homophenylalanine).

[0103] In embodiments, the cCPP of Formula (3) can include those having one of the following sequences: hFf-Nal-GrGr; bhF-F-Nal-SRSR; bhF-F-Nal-GRGR; bhF-F-Nal-HRHR; bhF-f-Nal-GrGr; bhF-f-Nal-SRSR; bhF-f-Nal-SrSr or bhFf-Nal-HrHr (where bhF – beta-homophenylalanine; hFf – homophenylalanine).

- 5 [0104] In embodiments, the cCPP of Formula (3) can include those having one of the following sequences: hFf-Nal-GrGrQ; β hF-F-Nal-SRSRQ; β hF-F-Nal-GRGRQ; β hF-F-Nal-HRHRQ; β hF-f-Nal-GrGrQ; β hF-f-Nal-SRSRQ; β hF-f-Nal-SrSrQ or β hFf-Nal-HrHrQ.

Linker

[0105] The EEV can include one or more linker arms. The linker can link a cargo to the cCPP.

- 10 The linker can link an EP to the cCPP. The linker can be attached to the side chain of an amino acid of the cCPP, and the cargo can be attached at a suitable position on the linker.

[0106] The linker can be any appropriate moiety which can conjugate a cCPP to one or more additional moieties, e.g., an exocyclic peptide (EP) and/or a cargo. Prior to conjugation (e.g., to the cCPP and/or one or more additional moieties), the linker has two or more functional groups,

- 15 each of which is independently capable of forming a covalent bond (e.g., to the cCPP and/or one or more additional moieties). If the cargo is an oligonucleotide, the linker can be covalently bound to the 5' end of the cargo or the 3' end of the oligonucleotide cargo. The linker can be covalently bound to the 5' end of the oligonucleotide cargo. The linker can be covalently bound to the 3' end of the oligonucleotide cargo. If the cargo is a peptide, the linker can be covalently bound to the N-terminus or the C-terminus of the peptide cargo. The linker can be covalently bound to the backbone (e.g., somewhere in the middle and not at a terminus or termini) of the oligonucleotide or peptide cargo. The linker can be any appropriate moiety that conjugates a cCPP described herein to a therapeutic moiety such as an oligonucleotide, peptide or small molecule.

- 25 [0107] The linker can be covalently bound to cargo at any suitable location on the cargo. The linker can be covalently bound to the 3' end of oligonucleotide cargo or the 5' end of an oligonucleotide cargo. The linker can be covalently bound to the N-terminus or the C-terminus of a peptide cargo. The linker can be covalently bound to the backbone of an oligonucleotide or a peptide cargo. The linker can be covalently bound to the cargo via a bonding group M'.

[0108] The linker can be bound to the side chain of an amino acid (AA_{sc}) on the cCPP, including for example, aspartic acid, glutamic acid, glutamine, asparagine, or lysine, or a modified side chain of glutamine or asparagine (e.g., a reduced side chain having an amino group). The linker can be bound to the side chain of lysine on the cCPP.

[0109] In embodiments, the linker can include one or more polyethylene glycol (PEG) components. Each (PEG) component can have, for example, from 0 to 12 repeat (PEG) units. A (PEG) unit refers to the group $-(CH_2CH_2O)-$ which can be repeated any number of times. In embodiments, a linker can include 1 (PEG) component. In embodiments, a linker can include 2 (PEG) components. In embodiments, a linker can include 3 (PEG) components. In embodiments, a linker can include 4 (PEG) components. In embodiments, a linker can include 5 (PEG) components. In embodiments, the number of repeat (PEG) units in each (PEG) component present in a linker can be the same number of repeat (PEG) units or can be different numbers of repeat (PEG) units. In embodiments, a linker can include multiple (PEG) components, where multiple (PEG) components can be separated by other moieties, such as one or more amino acid (AA) components and/or one or more hydrophobic components (X).

[0110] The linker can include one or more amino acid components (AA)a', (AA)b', and (AA)b'', which indicate an amino acid component comprising amino acid sequences of length a', b', or b''. For example, when a' is 2, (AA)a' is an amino acid component of two amino acid residues that are the same or different. In embodiments, where a linker arm includes more than one amino acid (AA) component (e.g., (AA)a', (AA)b', and (AA)b''), the amino acid residue or sequence of amino acid residues in each amino acid (AA) component can be the same or different.

[0111] While not wishing to be bound by theory, it is believed that the linkers can influence efficacy and tolerability of the resultant construct, for example to increase efficacy. In embodiments, efficacy is increased without decreasing tolerability. In embodiments, one or more hydrophobic components (X) are added to a linker of the EEV. In embodiments, the length of the linker can be varied. The linker length can influence efficacy and tolerability. In some instances, as linker length decreases, efficacy increases and tolerability decreases; and as linker length increases, efficacy decreases and tolerability increases. In embodiments, increasing hydrophobicity of the linker can increase efficacy. In embodiments, the linker can include an amino acid component (AA)a', wherein a' is an integer from 0-12, an amino acid component (AA)b', wherein b' is an integer from 0-12, an amino acid component (AA)b'', wherein b'' is an

integer from 0-12, a polyethylene glycol (PEG)x' component, wherein x' is an integer from 0-12, a polyethylene glycol (PEG)z' component, wherein z' is an integer from 0-12, a polyethylene glycol (PEG)z'' component, wherein z'' is an integer from 0-12, one or more hydrophobic components (e.g., X^{o'}, X^{o''}, etc.), or a combination thereof.

5 [0112] A hydrophobic component X can be added at one or more positions on the linker. For example, X can be present in a linker between the CPP and EP; or in a linker between the CPP and the cargo. In embodiments, X can be the bonding group between the CPP and EP. In embodiments, X is not the bonding group between the CPP and EP. In embodiments, X can be the bonding group between the CPP and cargo. In embodiments, X is not the bonding group
10 between the CPP and cargo. X can form part of the backbone of the linker between the CPP and EP. X can form part of the backbone of the linker between the CPP and cargo. X can be appended to the linker. X can be appended to the side chain of a lysine residue in the linker. X can be appended to the linker between the CPP and EP. X can be appended to the linker between the CPP and cargo.

15 [0113] In embodiments, the hydrophobic component (X) in the linker can be aliphatic, alkene, alkyne, aromatic (e.g., carbocyclic or heteroaromatic), or a combination of aliphatic and aromatic.

[0114] X can be a D or L amino acid residue with a hydrophobic side chain. X can be a naturally occurring or a non-naturally occurring amino acid residue with a hydrophobic side chain. X can
20 be an amino acid residue with an aromatic side chain. X can be an amino acid residue with a heteroaromatic side chain. X can be selected from phenylalanine, 3-(4',4-biphenyl)-L-alanine, tryptophan, tyrosine, valine, isoleucine, leucine, or histidine, or a combination thereof. X can be 2-naphthylalanine. X can be Nal. X can be d-Nal (nal). X can be 3-(4',4-biphenyl)-L-alanine. X can be Bip. X can be D-Bip (bip). X can be a C4-C8 alkyl hydrocarbon. X can be a C6 alkyl
25 hydrocarbon.

[0115] In embodiments, the hydrophobic component X comprises an optionally substituted alkyl group. The alkyl group can comprise a branched alkyl group. In embodiments, the alkyl group comprises a double bond. In embodiments, the alkyl group comprises an ethyl or propyl group. In embodiment, the alkyl group is a butyl, pentyl, or hexyl group. In embodiment, the alkyl
30 group is -CH₂(CH₂)_nCH₂-, CH₃(CH₂)_nCH₂-, C(H)C(CH₂)_nCH₂-, CH₃(CH₂)_nCH₂NH-, C(H)C(CH₂)_nCH₂NH-, CH₃(CH₂)_nCO-, CH₃(CH₂)_nCH₂O-, CH₃(CH₂)_nCH₂S-, -

CH₂(CH₂)_nCH₂SH-, -OC(CH₂)_nCH₂-, -OC(CH₂)_nCH₂NH-, -OC(CH₂)_nCO-, -OC(CH₂)_nCH₂O-, -OC(CH₂)_nCH₂S-, -HNC(CH₂)_nCH₂-, -HNC(CH₂)_nCH₂NH-, -HNC(CH₂)_nCO-, -HNC(CH₂)_nCH₂O-, -HNC(CH₂)_nCH₂S-, -OCH₂(CH₂)_nCH₂-, -OCH₂(CH₂)_nCH₂NH-, -OCH₂(CH₂)_nCO-, -OCH₂(CH₂)_nCH₂O-, -OCH₂(CH₂)_nCH₂S-, NH₂(CH₂)_nCH₂-, SH(CH₂)_nCH₂-, or N₃-C(O)CH₂(CH₂)_nCH₂-, group, wherein “n” is 4-34, inclusive.

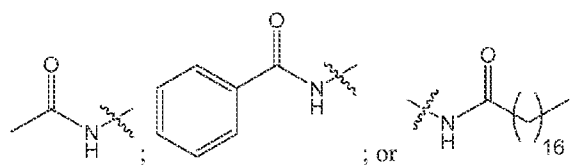
[0116] In embodiments, the hydrophobic component X can be an optionally substituted aromatic group. Aromatic groups can be carbocyclic heteroaromatic, monocyclic, bicyclic or polycyclic. Carbocyclic aryl groups include benzene, naphthalene, phenanthrene, phenol, aniline, anthracene and the like. Heteroaryl groups include, for example, pyrrole, furan, thiophene, imidazole, oxazole, thiazole, triazole, pyrazole, pyridine, pyrazine, pyridazine and pyrimidine, and the like.

[0117] In embodiments, the hydrophobic component X may comprise one or more hydrophobic amino acid residues. The amino acid residues can be D or L. The amino acid residues can be natural hydrophobic amino acid or non-natural hydrophobic amino acid. In embodiments, the hydrophobic amino acid residues comprise a substituted or unsubstituted alkyl, alkenyl, alkynyl, aryl or aralkyl side chain wherein the alkyl, alkenyl and alkynyl side chain includes at most one heteroatom for every six carbon atoms. X can be a D or L amino acid residue with a hydrophobic side chain. X can be a naturally occurring or a non-naturally occurring amino acid residue with a hydrophobic side chain. X can be an amino acid residue with an aromatic side chain. X can be an amino acid residue with a heteroaromatic side chain. The hydrophobic component X can comprise an amino acid residue with a hydrophobic side chain can be selected from valine, proline, alanine, leucine, isoleucine, phenylalanine, cysteine, glycine, histidine, and methionine and tryptophan. The amino acid residue with a hydrophobic side chain can be selected from the group consisting of glycine, phenylglycine, alanine, valine, leucine, isoleucine, norleucine, phenylalanine, tryptophan, naphthylalanine, proline, and combinations thereof, wherein the aromatic side chains on phenylglycine, phenylalanine, tryptophan, or naphthylalanine are each optionally substituted. The amino acid residue with a hydrophobic side chain can be naphthylalanine, lysine-naphthylalanine, biphenylalanine, or lysine-biphenylalanine. X can be an aliphatic hydrocarbon. X can be a C₄-C₁₂ aliphatic hydrocarbon. X can be a C₄-C₈ aliphatic hydrocarbon. X can be a C₆ aliphatic hydrocarbon. X can be selected from phenylalanine, 3-(4',4'-biphenyl)-L-alanine, tryptophan, tyrosine, valine, isoleucine, leucine, or histidine, or a combination thereof. X can be amino acid

selected from tryptophan, tyrosine, isoleucine, leucine, histidine, phenylalanine, or a combination thereof.

[0118] X can be 2-naphthylalanine. X can be Nal. X can be d-Nal (nal). X can be 3-(4',4-biphenyl)-L-alanine (Bip). X can be 3-(4',4-biphenyl)-D-alanine (d-Bip or bip).

- 5 [0119] In embodiments, the hydrophobic component X can include amino acid subunits that have been modified. For example, the amino terminal or carboxy terminal amino acid subunit may be modified. Such modifications include capping the amino terminus or the carboxy terminus with a group making the amino acid subunit more hydrophobic. For example, the amino terminus may be capped with an acyl group (e.g., acetyl, benzoyl, or stearoyl moiety). For example, the
10 amino terminus of the of the modified amino acid subunit can be depicted as follows:



[0120] In embodiments, X can be appended to the backbone of the linker via lysine or a $\text{SH}(\text{CH}_2)_n\text{CH}_2-$, or $\text{N}_3-\text{C}(\text{O})\text{CH}_2(\text{CH}_2)_n\text{CH}_2-$.

- [0121] In embodiments, the size of an aromatic or heteroaromatic group present in the
15 hydrophobic component X may be selected to improve cytosolic delivery efficiency of the Cargo. While not wishing to be bound by theory, it is believed that the presence of a hydrophobic component X in the linker of the EEV may improve cytosolic delivery efficiency of the Cargo as compared to an otherwise identical EEV that does not include the hydrophobic component X.

- 20 [0122] The linker can comprise hydrocarbon linker.

[0123] The linker can comprise a cleavage site. The cleavage site can be a disulfide, or caspase-cleavage site (e.g., Val-Cit-PABC).

- [0124] The linker can comprise: (i) one or more D or L amino acids, each of which is optionally substituted; (ii) one or more $-(\text{R}^1\text{-J-R}^2)_i-$ subunits, wherein each of R^1 and R^2 , at each instance,
25 are independently selected from alkylene, each J is independently C, NR^3 , $-\text{NR}^3\text{C}(\text{O})-$, S, and O, wherein R^3 is independently selected from H, alkyl, alkenyl, alkynyl, carbocyclyl, and heterocyclyl, each of which is optionally substituted, and i is an integer from 1 to 50.

[0125] The linker can comprise one or more D or L amino acids and/or $-(\text{OCH}_2\text{CH}_2)_i-$ i is an integer from 0 to 60; or combinations thereof. i can be an integer from 0 to 12. i can be 0, 2, 4, 8

or 12. i can be 0. i can be 2. i can be 4. i can be 8. i can be 12. “ $-(\text{OCH}_2\text{CH}_2)_i$ ” can also be referred to as polyethylene glycol (PEG).

[0126] The linker can comprise one or more amino acids. The linker can comprise one or more amino acid components $(\text{AA})_{a'}$, $(\text{AA})_{b'}$, and/or $(\text{AA})_{b''}$ comprising from 1 to 12 amino acids, or 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, or 12 amino acids. The amino acid component can comprise from 1 to 10 amino acids. The amino acid component can comprise from 1 to 8 amino acids. The amino acid component can comprise from 2-5 amino acids. The linker can comprise one or more PEG components $-(\text{OCH}_2\text{CH}_2)_i$, wherein i is an integer from 0 to 12. The PEG components can be represented as $(\text{PEG})_{x'}$, $(\text{PEG})_{z'}$, and/or $(\text{PEG})_{z''}$. The linker can comprise one or more hydrophobic components $(X^{o'})$ and $(X^{o''})$. The hydrophobic component can be appended to a side chain of an amino acid in the linker. The hydrophobic component $(X^{o'})$ can be appended to a side chain of lysine in linker $(\text{K}(X^{o'}))$. The linker can comprise a bonding group (M).

[0127] The linker can comprise (i) a β alanine residue and lysine residue; (ii) $-(\text{J}-\text{R}^1)_i$; or (iii) a combination thereof. Each R^1 can independently be alkylene, alkenylene, alkynylene, carbocyclyl, or heterocyclyl, each J is independently C, NR^3 , $-\text{NR}^3\text{C}(\text{O})-$, S, or O, wherein R^3 is H, alkyl, alkenyl, alkynyl, carbocyclyl, or heterocyclyl, each of which is optionally substituted, and z'' can be an integer from 1 to 50. Each R^1 can be alkylene and each J can be O.

[0128] The linker can comprise (i) residues of β -alanine, glycine, lysine, 4-aminobutyric acid, 5-aminopentanoic acid, 6-aminohexanoic acid or combinations thereof; and (ii) $-(\text{R}^1\text{-J})_{i'}$ or $-(\text{J}-\text{R}^1)_{i'}$.

Each R^1 can independently be alkylene, alkenylene, alkynylene, carbocyclyl, or heterocyclyl, each J is independently C, NR^3 , $-\text{NR}^3\text{C}(\text{O})-$, S, or O, wherein R^3 is H, alkyl, alkenyl, alkynyl, carbocyclyl, or heterocyclyl, each of which is optionally substituted, and i' can be an integer from 1 to 50. Each R^1 can be alkylene and each J can be O. The linker can comprise glycine, beta-alanine, 4-aminobutyric acid, 5-aminopentanoic acid, 6-aminohexanoic acid, or a combination thereof.

[0129] The linker can also incorporate a cleavage site, including a disulfide $[\text{NH}_2-(\text{CH}_2\text{O})_n-\text{S}-\text{S}-(\text{CH}_2\text{O})_n-\text{COOH}]$, or caspase-cleavage site (Val-Cit-PABC).

[0130] The linker can include a residue of glycine or β -alanine.

[0131] The linker can be bivalent and link the cCPP to a cargo. The linker can be bivalent and link the cCPP to an exocyclic peptide (EP).

[0132] The linker can be trivalent and link the cCPP to a cargo and to an EP.

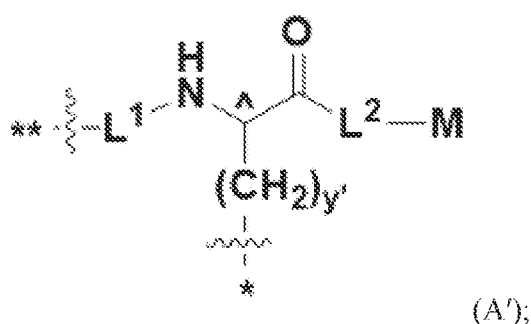
[0133] The linker can be a bivalent or trivalent C₁-C₅₀ alkylene, wherein 1-25 methylene groups are optionally and independently replaced by -N(H)-, -N(C₁-C₄ alkyl)-, -N(cycloalkyl)-, -O-, -C(O)-, -C(O)O-, -S-, -S(O)-, -S(O)₂-, -S(O)₂N(C₁-C₄ alkyl)-, -S(O)₂N(cycloalkyl)-, -N(H)C(O)-, -N(C₁-C₄ alkyl)C(O)-, -N(cycloalkyl)C(O)-, -C(O)N(H)-, -C(O)N(C₁-C₄ alkyl), -

5 C(O)N(cycloalkyl), aryl, heterocyclyl, heteroaryl, cycloalkyl, or cycloalkenyl. The linker can be a bivalent or trivalent C₁-C₅₀ alkylene, wherein 1-25 methylene groups are optionally and independently replaced by -N(H)-, -O-, -C(O)N(H)-, or a combination thereof.

[0134] The cargo can be coupled to the glutamic acid of the cyclic peptide, which converts the glutamic acid to glutamine. The linker (L) can couple the cargo to the glutamine/glutamic acid of
10 the cyclic peptide. In embodiments, a linker (L) is covalently bound to the backbone of the cargo.

[0135] The linker can be a trivalent linker with the structure shown in Formula (A'):

Formula (A')



15 wherein:

** is a point of attachment to a linear exocyclic peptide (EP) as defined herein;

* is a point of attachment to a cell penetrating peptide (CPP) as defined herein;

L¹ and L², are, independently, a linker arm;

^ indicates L- or D-stereochemistry;

20 y' is an integer from 1 to 5; and

M comprises a reactive handle.

[0136] In embodiments, L¹ and L², are each, independently, absent, or comprise hydrocarbon component (e.g., NRH-(CH₂)_n-COOH), a polyethylene glycol (PEG) component, a hydrophobic component (X), an amino acid (AA) component comprising one or more amino acid residues, or
25 a combination thereof. In embodiments, L¹ comprises a hydrocarbon linker (e.g., NRH-(CH₂)_n-COOH). In embodiments, L¹ comprises a polyethylene glycol (PEG)_{x'} linker, wherein x' is an

integer from 1-12. In embodiments, L^1 comprises an amino acid component $(AA)a'$, wherein a' is an integer from 1-12. In embodiments, L^2 comprises a hydrocarbon component (e.g., $NRH-(CH_2)_n-COOH$). In embodiments, L^2 comprises one or more polyethylene glycol components (e.g., $(PEG)z'$ and/or $(PEG)z''$, wherein z' and z'' are, independently, integers from 1-12). In

5 embodiments, L^2 comprises one or more hydrophobic components ($X^{o'}$, $X^{o''}$, etc.). In embodiments, L^2 comprises one or more amino acid components comprising one or more amino acid residues $(AA)b'$ and/or $(AA)b''$, wherein b' and b'' are, independently, an integer from 1-12.

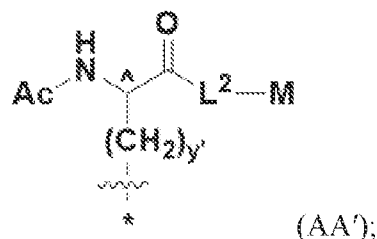
[0137] In embodiments, L^1 , L^2 or cCPP comprise one or more hydrophobic component(s) (X).

[0138] In embodiments, L^1 comprises one or more amino acid (AA) components, one or more

10 PEG components, or a combination thereof, and L^2 comprises one or more amino acid (AA) components, one or more PEG components, one or more hydrophobic components (X), or a combination thereof. In embodiments, L^1 comprises one or more $(AA)a'$ and $(PEG)x'$ components, or a combination thereof. In embodiments, L^2 comprises one or more $(AA)b'$, $(AA)b''$, $(PEG)z'$, $(PEG)z''$, hydrophobic components ($X^{o'}$, $X^{o''}$, etc.), or a combination thereof.

15 [0139] In embodiments, the linker comprises Formula (AA'):

Formula (AA'):



wherein

* is a point of attachment to a cell penetrating peptide (CPP) as defined herein;

20 Ac is acetyl;

L^2 is a linker arm comprising one or more $(AA)b'$, $(AA)b''$, $(PEG)z'$, and/or $(PEG)z''$, one or more hydrophobic groups ($X^{o'}$) or ($X^{o''}$), or a combination thereof;

$(AA)b'$, and $(AA)b''$, are, independently, an amino acid linker comprising at least one amino acid residue;

25 b' and b'' are, independently, an integer from 0 to 12;

z' and z'' are, independently, an integer from 0 to 12;

^ indicates L- or D-stereochemistry;

M is a reactive handle; and

y' is an integer from 1 to 5.

[0140] In embodiments, the hydrophobic group $X^{o'}$, can be appended to a side chain of a lysine residue (K).

5 [0141] In embodiments, L^1 comprises (AA)a', and/or (PEG)x'. In embodiments, L^1 comprises (AA)a' and (PEG)x'. In embodiments, L^1 comprises (AA) a'. In embodiments, L^1 comprises (PEG)x'. In embodiments, L^1 comprises (AA)a'. In embodiments, (AA)a' can be appended from a side chain of a lysine amino acid residue. In embodiments, L^1 comprises at least one (AA)a'. In
10 embodiments, L^1 comprises at least one (PEG)x'. In embodiments, L^1 comprises one (AA)a'. In
embodiments, L^1 least one (PEG)x'.

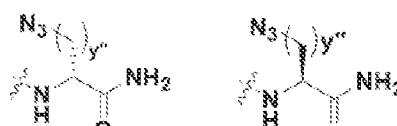
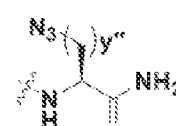
[0142] In embodiments, L^1 is absent. In embodiments, L^1 is absent, and L^2 comprises (AA)b', (AA)b'', (PEG)z', (PEG)z'', one or more hydrophobic components (e.g., $X^{o'}$, $X^{o''}$), or a combination thereof. In embodiments, when L^1 is absent, the lysine linked to the cyclic peptide is acylated (Ac).

15 [0143] In embodiments, L^2 comprises (AA)b', (AA)b'', (PEG)z', (PEG)z'', one or more a hydrophobic components ($X^{o'}$), ($X^{o''}$) or a combination thereof. In embodiments, L^2 comprises (AA)b' and (PEG)z'. In embodiments, L^2 comprises (AA)b' and a hydrophobic component ($X^{o'}$). In embodiments, L^2 comprises (PEG)z' and a hydrophobic component ($X^{o'}$). In embodiments, L^2 comprises (AA)b'. In embodiments, (AA)b' can be appended from a side chain of a lysine amino
20 acid residue. In embodiments, L^2 comprises (PEG)z'. In embodiments, L^2 comprises a hydrophobic component ($X^{o'}$). In embodiments, the hydrophobic component ($X^{o'}$) is appended from a side chain of a lysine amino acid residue. In embodiments, L^2 comprises (AA)b' and (AA)b''. In
embodiments, L^2 comprises (PEG)z' and (PEG)z''. In embodiments, L^2 comprises at least one hydrophobic component ($X^{o'}$), and/or ($X^{o''}$). In embodiments, L^2 comprises two amino acid
25 components (AA)b' and (AA)b''. In embodiments, the two amino acid components (AA)b' and (AA)b'' are different. In embodiments, the two amino acid components (AA)b' and (AA)b'' are the same. In embodiments, the two amino acid components (AA)b' and (AA)b'' are separated by a PEG component (PEG)z'. In embodiments, the two amino acid components (AA)b' and (AA)b''
are separated by a hydrophobic component ($X^{o'}$). In embodiments, L^2 comprises two PEG
30 components (PEG)z' and (PEG)z''. In embodiments, the two PEG components (PEG)z' and (PEG)z'' are the same. In embodiments, the two PEG components (PEG)z' and (PEG)z'' are

different. In embodiments, the two PEG components (PEG)_{z'} and (PEG)_{z''} are separated by an amino acid component (AA)_{b'} or (AA)_{b''}. In embodiments, the two PEG components (PEG)_{z'} and (PEG)_{z''} are separated by a hydrophobic component (e.g., X^{o'}, X^{o''}). In embodiments, L² comprises two hydrophobic components (e.g., X^{o'}, X^{o''}). In embodiments, the two hydrophobic components (X^{o'}, X^{o''}) are different. In embodiments, the two hydrophobic components (X^{o'}, X^{o''}) are the same. In embodiments, the two hydrophobic components (X^{o'}, X^{o''}) are separated by a PEG component. In embodiments, the two hydrophobic components (X^{o'}, X^{o''}) are separated by an amino acid (AA) component.

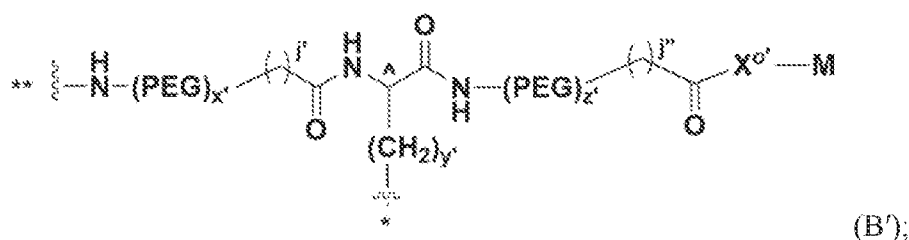
[0144] M comprises a functional group (referred to herein as a reactive handle) that can react with a corresponding functional group on a cargo to form an EEV-cargo conjugate or a linker-cargo conjugate. Following conjugation, the EEV-cargo or linker-cargo conjugate includes a bonding group M', which is the reaction product of the two functional groups.

[0145] In embodiments, M can be -OH (e.g., as part of the C terminus of an amino acid). In

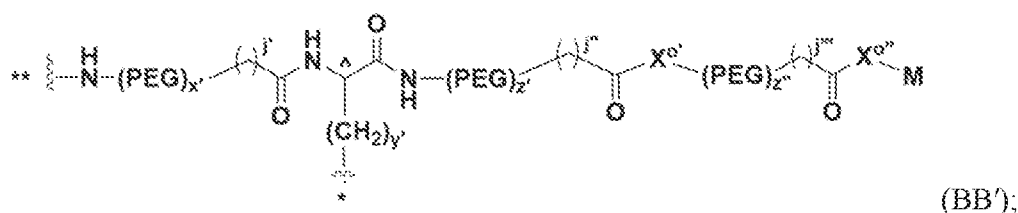
embodiments, M comprises -OH, , or , herein y'' is an integer from 1-4. y'' can be 1. y'' can be 2. y'' can be 3. y'' can be 4.

[0146] In embodiments, the linker Formula (A') can have a Formula selected from Formula (B'), (BB'), (C'), (D'), (E'), or (F'):

Formula (B'):



Formula (BB'):



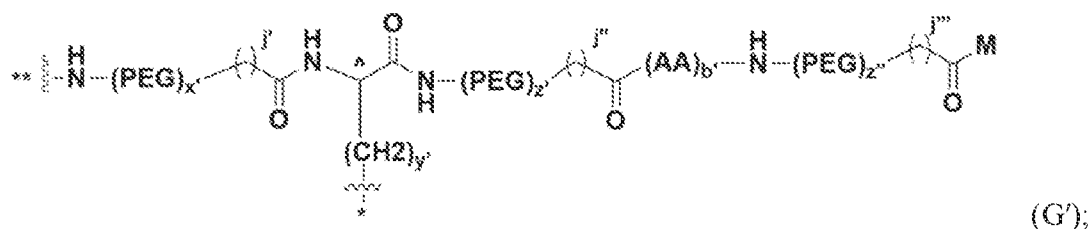
(AA) b' and (AA) b'' are each independently an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

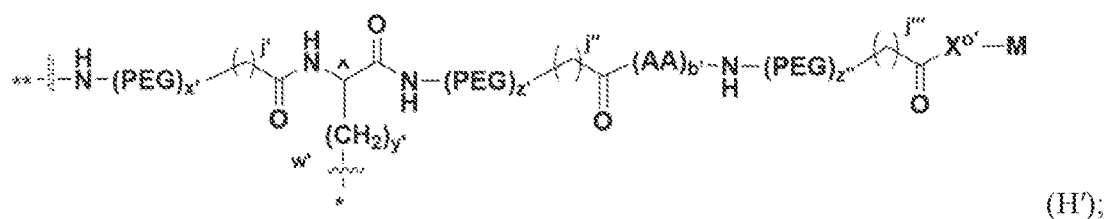
b'' is an integer from 0 to 12.

- 5 [0147] In embodiments, the linker Formula (A') can have a Formula selected from Formula (G'), (H'), (I'), (J'), (K'), (L'), or (M'):

Formula (G'):

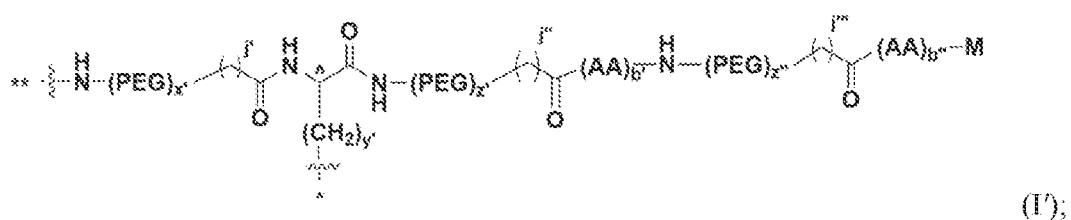


Formula (H'):

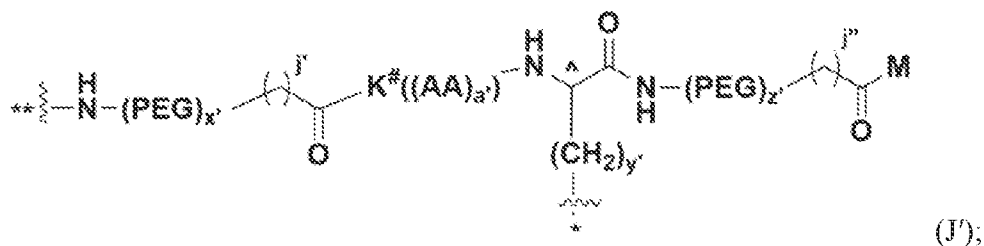


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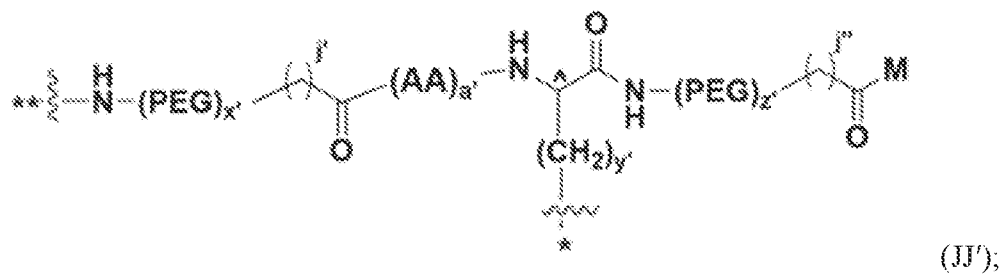
Formula (I'):



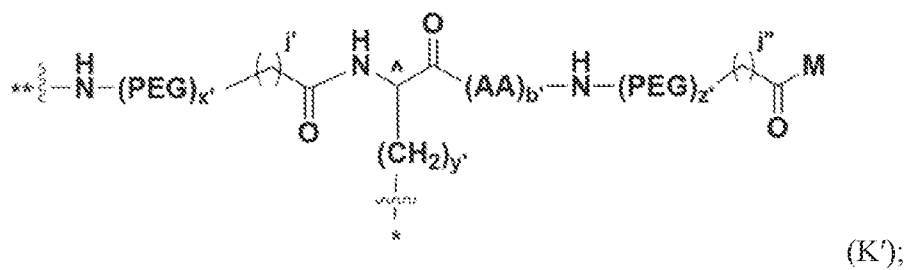
Formula (J'):



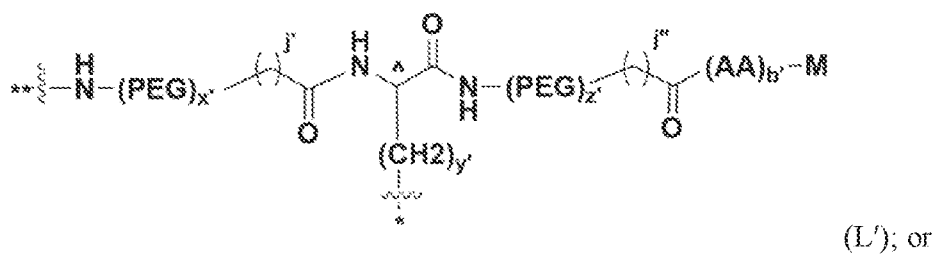
Formula (JJ'):



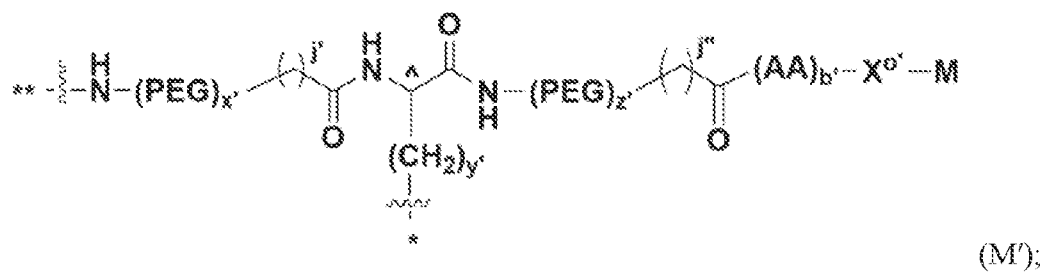
Formula (K'):



5 Formula (L'):



Formula (M'):



wherein:

- 10 x' is an integer from 1 to 12;
 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;
 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1 or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ is a hydrophobic component;

5 $K^\#$ is D-lysine or L-lysine residue;

$(AA)a'$, $(AA)b'$, and $(AA)b''$ are each independently an amino acid (AA) component comprising at least one amino acid residue;

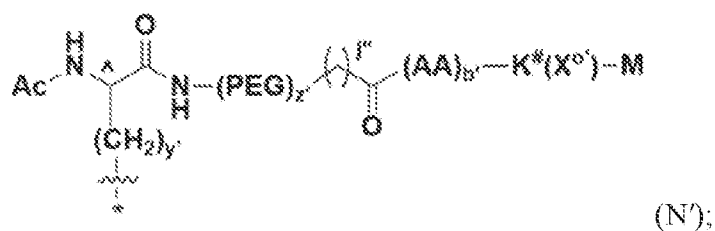
a' is an integer from 0 to 12;

b' is an integer from 0 to 12; and

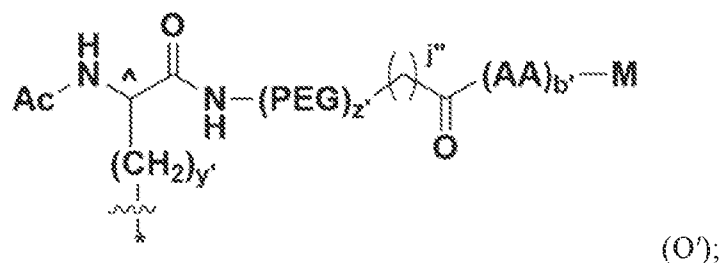
10 b'' is an integer from 0 to 12.

[0148] In embodiments, the linker Formula (AA') can have a Formula selected from Formula (N'), (O), (P'), or (Q'):

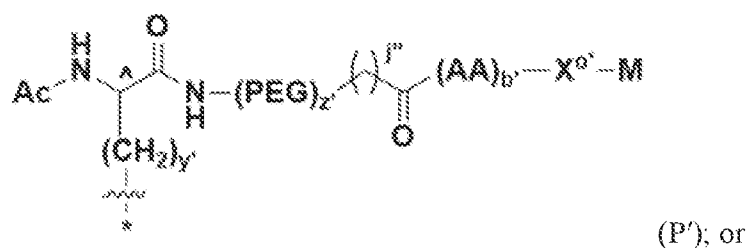
Formula (N'):



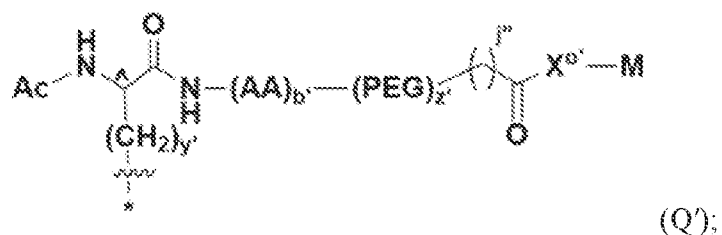
15 Formula (O'):



Formula (P'):



Formula (Q):



wherein:

z' is an integer from 0 to 12;

5 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$\text{X}^{\text{O}'}$ is a hydrophobic component;

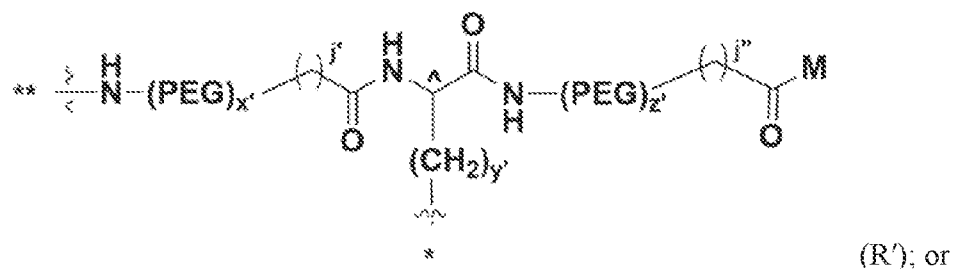
$\text{K}^{\#}$ is D-lysine or L-lysine residue;

$(\text{AA})_{b'}$ is an amino acid (AA) component comprising at least one amino acid residue; and

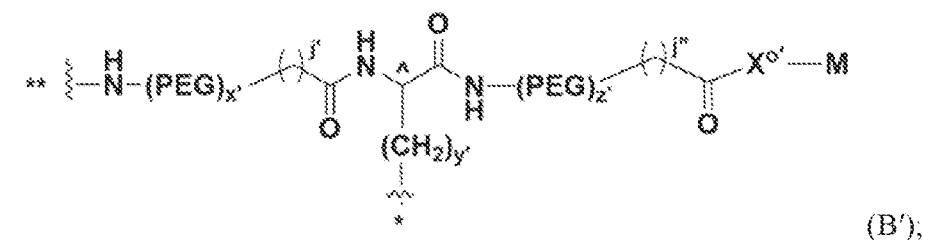
b' is an integer from 0 to 12.

10 [0149] In embodiments, the linker Formula (A') can have a Formula selected from Formula (R') or (B'):

Formula (R'):



Formula (B'):



wherein:

x' is an integer from 1 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0; and

$X^{o'}$ is a hydrophobic component.

[0150] With the exception of glycine (G), which has H as a side chain, amino acids each have an asymmetric or chiral α -carbon atom and exists in two enantiomeric forms, designated “D-” and

5 “L-”. The “L-” isomers are most commonly found in naturally occurring proteins. While not wishing to be bound by theory, it is believed that the chirality of the amino acids in a cell-penetrating peptide (CPP) or endosomal escape vehicle (EEV) can impact cytosolic delivery efficiency, as well as toxicity. Disclosed herein are CPP and EEV constructs in which the stereochemistry of the amino acids within the peptide sequences of the construct has been
10 modified to influence cytosolic delivery efficiency and/or reduce toxicity.

[0151] In embodiments, the linker comprises a structure selected from Formula (B'), (BB'), (C'), (D'), (E'), or (F'), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AA_{sc} conjugates cCPP to the linker.

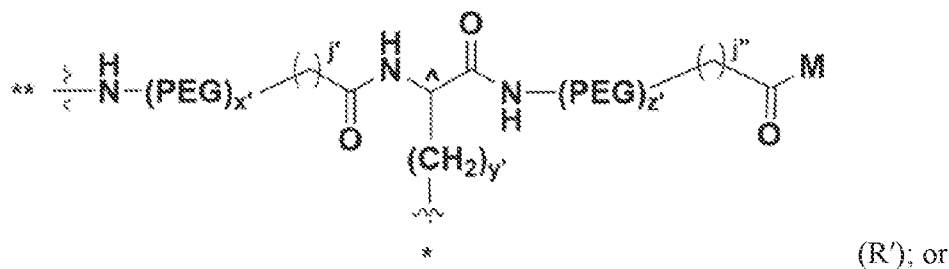
15 [0152] In embodiments, the linker comprises a structure selected from Formula (G'), (H'), (I'), (J'), (JJ'), (K'), (L'), or (M'), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AA_{sc} conjugates cCPP to the linker.

[0153] In embodiments, the linker comprises a structure selected from Formula (N'), (O'), (P'), or
20 (Q'), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

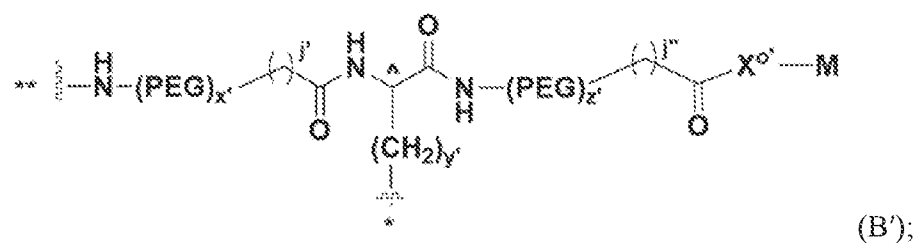
[0154] In embodiments, the linker comprises a structure selected from Formula (R') or (B'), wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids
25 and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker

[0155] In embodiments, the linker Formula (A') can have a Formula selected from Formula (R'), or (B'):

Formula (R'):



Formula (B'):



5 wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0; and

10 X^{o'} comprises a hydrophobic component,

wherein the EP comprises all D-amino acid residues; and the cCPP comprises D-amino acid residues, achiral amino acids and AAsc, wherein AAsc is an amino acid residue side chain that conjugates the cCPP to the linker.

[0156] In embodiments, the cCPP comprises at least 2 D-amino acid residues with a

15 hydrophobic side chain. In embodiments, the cCPP comprises 2 D-amino acid residues with a

hydrophobic side chain. In embodiments, the cCPP comprises 3 D-amino acid residues with a

hydrophobic side chain. In embodiments, the cCPP comprises at least 2 D-arginine amino acid

residues. In embodiments, the cCPP comprises 2 D-arginine amino acid residues. In

embodiments, the cCPP comprises 3 D-arginine amino acid residues. In embodiments, the cCPP

20 comprises 4 D-arginine amino acid residues. In embodiments, the cCPP comprises 5 D-arginine

amino acid residues. In embodiments, the cCPP comprises 6 D-arginine amino acid residues. In

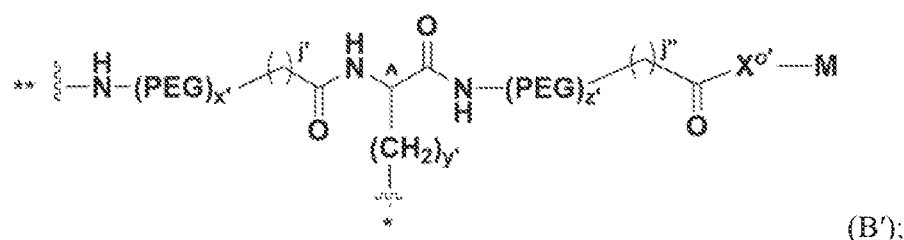
embodiments, the cCPP comprises at least 2 D-phenylalanine residues. In embodiments, the

cCPP comprises 2 D-phenylalanine residues. In embodiments, the cCPP comprises 3 D-

phenylalanine residues. In embodiments, the cCPP comprises at least 2 glycine residues. In embodiments, the cCPP comprises 2 glycine residues. In embodiments, the cCPP comprises 3 glycine residues. In embodiments, the cCPP comprises 4 glycine residues. In embodiments, the cCPP comprises 5 glycine residues. In embodiments, the cCPP comprises 6 glycine residues. In 5 embodiments, the cCPP comprises at least 2 D-amino acid residues with a hydrophobic side chain and at least 2 D-arginine residues. In embodiments, the cCPP comprises at least 2 D-phenylalanine residues and at least 2 D-arginine residues. In embodiments, AA_{SC} is a side chain of L-glutamine.

[0157] The linker can be of Formula (B'):

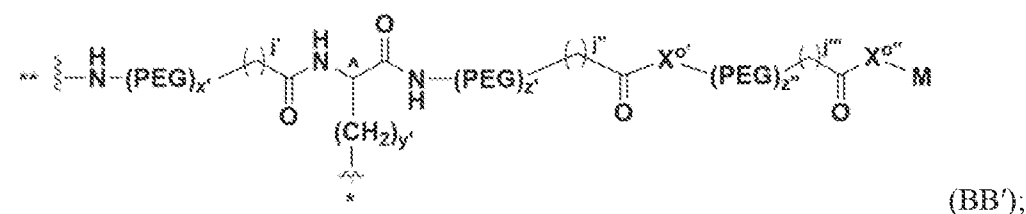
10 *Formula (B')*:



wherein **, *, x', j', ^, y', z', j'', X^{o'}, and M are as defined herein.

[0158] The linker can be of Formula (BB'):

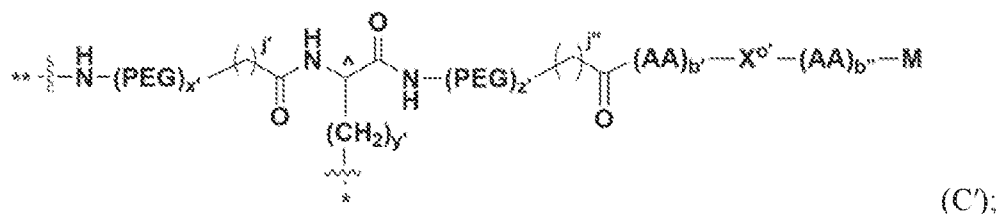
Formula (BB'):



wherein **, *, x', j', ^, y', z', j'', z'', j''', X^{o'}, X^{o''}, and M are as defined herein.

[0159] The linker can be of Formula (C'):

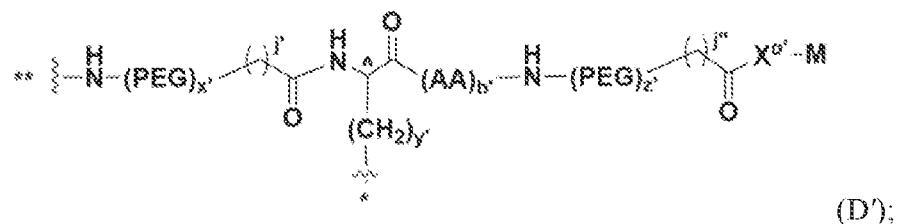
Formula (C'):



wherein **, *, x', j', ^, y', z', j'', X^{o'}, (AA)b', (AA)b'', b', b'', and M are as defined herein.

[0160] The linker can be of Formula (D'):

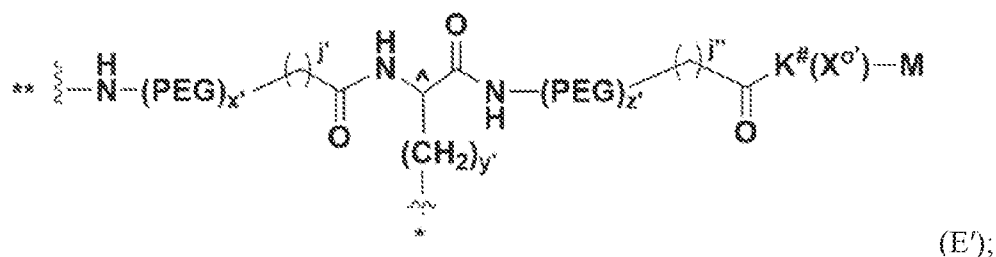
Formula (D'):



5 wherein **, *, x', j', ^, y', z', j'', X^{o'}, (AA)b', b', and M are as defined herein.

[0161] The linker can be of Formula (E'):

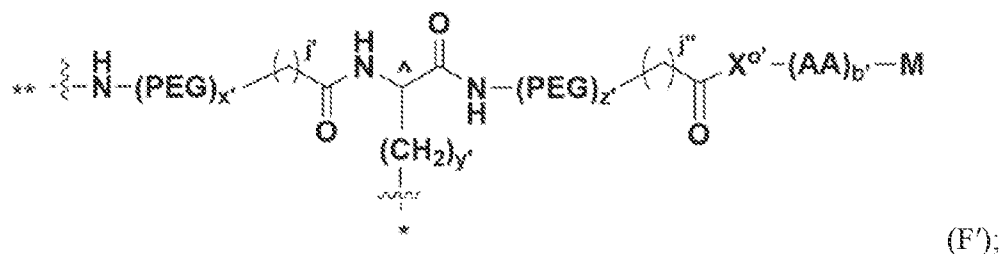
Formula (E'):



wherein **, *, x', j', ^, y', z', j'', X^{o'}, K[#], and M are as defined herein.

10 [0162] The linker can be of Formula (F'):

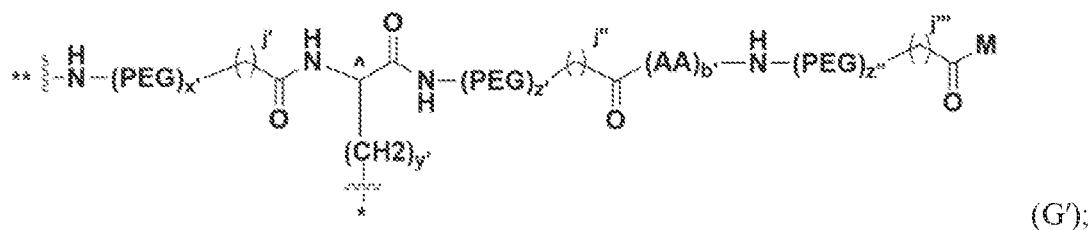
Formula (F'):



[0163] wherein **, *, x', j', ^, y', z', j'', X^{o'}, (AA)b', b', and M are as defined herein.

[0164] The linker can be of Formula (G'):

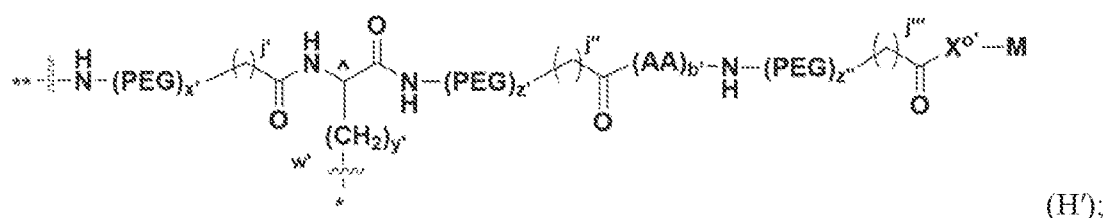
Formula (G'):



wherein **, *, x', j', ^, y', z', j'', z'', j''', (AA)b', b', and M are as defined herein.

[0165] The linker can be of Formula (H'):

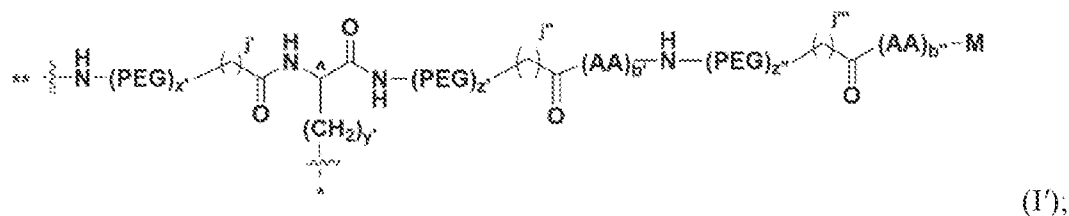
5 Formula (H'):



wherein **, *, x', j', ^, y', z', j'', z'', j''', (AA)b', b', X^{o'}, and M are as defined herein.

[0166] The linker can be of Formula (I'):

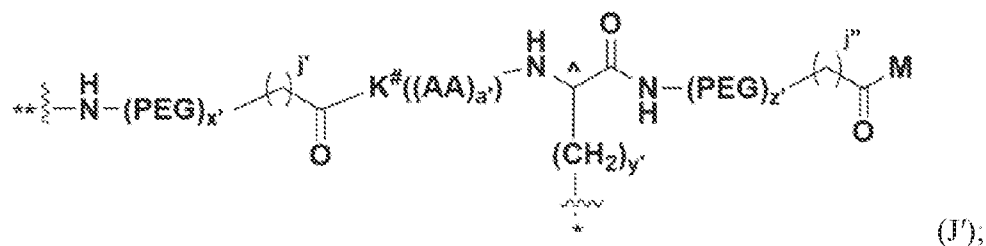
Formula (I'):



wherein **, *, x', j', ^, y', z', j'', z'', j''', (AA)b', (AA)b'', b', b'', and M are as defined herein.

[0167] The linker can be of Formula (J'):

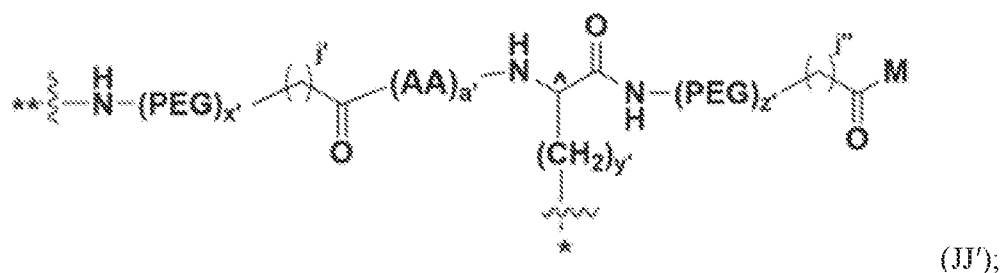
Formula (J'):



wherein **, *, x', j', K[#], (AA)a', a', ^, y', z', j'', and M are as defined herein.

[0168] The linker can be of Formula (JJ'):

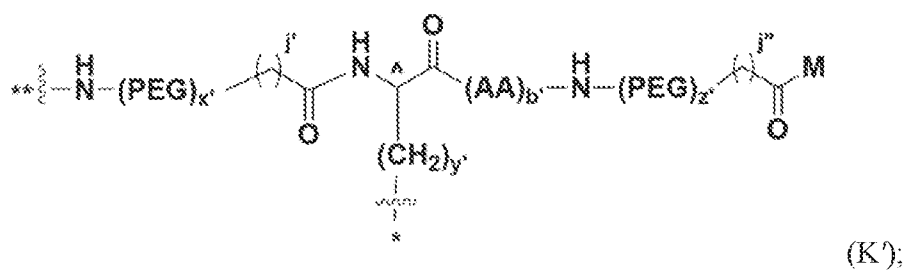
Formula (JJ'):



wherein **, *, x', j', (AA)_{a'}, a', ^, y', z', j'', and M are as defined herein.

[0169] The linker can be of Formula (K'):

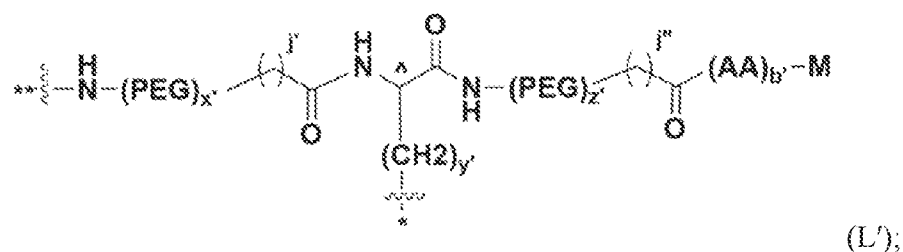
5 Formula (K'):



wherein **, *, x', j', ^, y', z', j'', (AA)_{b'}, b', and M are as defined herein.

[0170] The linker can be of Formula (L'):

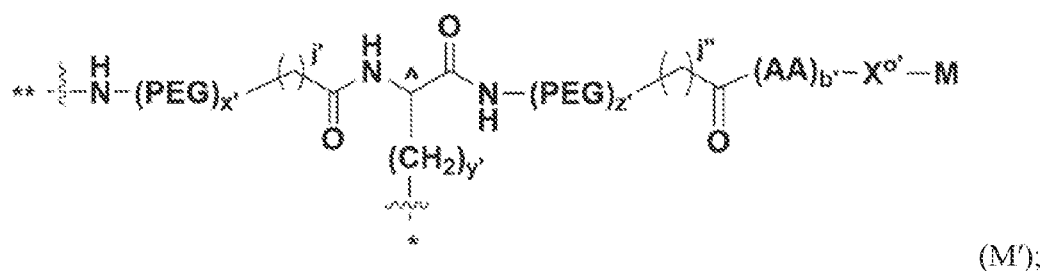
Formula (L'):



10 wherein **, *, x', j', ^, y', z', j'', (AA)_{b'}, b', and M are as defined herein.

[0171] The linker can be of Formula (M'):

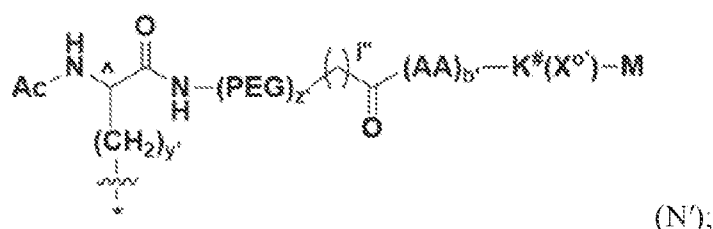
Formula (M'):



wherein^{**}, *, x', j', ^, y', z', j'', (AA)b', b', X^{o'}, and M are as defined herein.

[0172] The linker can be of Formula (N'):

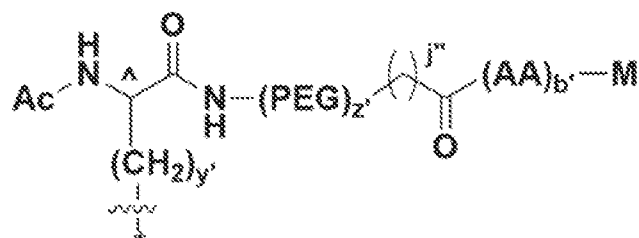
5 *Formula (N')*:



wherein Ac, *, ^, y', z', j'', (AA)b', b', K[#], X^{o'}, and M are as defined herein.

[0173] The linker can be of Formula (O'):

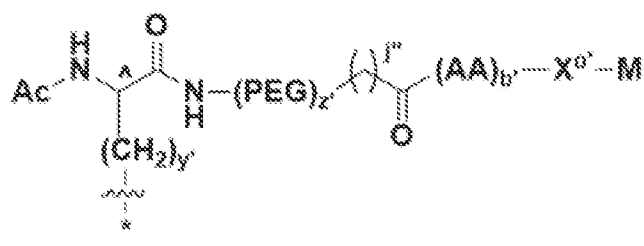
Formula (O'):



wherein Ac, *, ^, y', z', j'', (AA)b', b', and M are as defined herein.

[0174] The linker can be of Formula (P'):

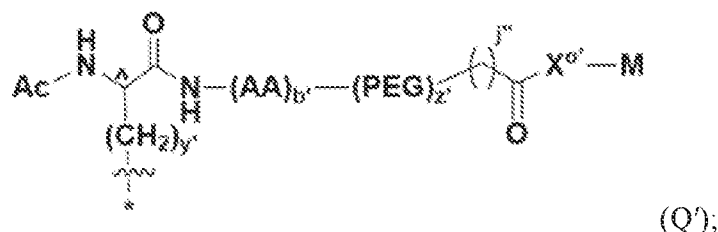
Formula (P'):



wherein Ac, *, ^, y', z', j'', (AA)b', b', X⁰', and M are as defined herein.

[0175] The linker can be of Formula (Q'):

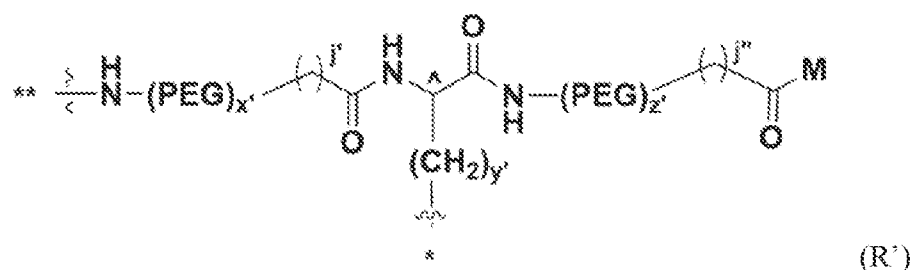
Formula (Q'):



wherein Ac, *, ^, y', z', j'', (AA)b', b', X^{o'}, and M are as defined herein.

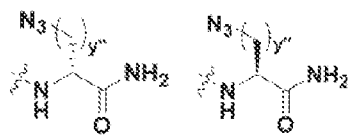
5 [0176] The linker can be of Formula (R'):

Formula (R'):



wherein *, **, x', j', ^, y', z', j'', and M are as defined herein.

[0177] In embodiments wherein L² comprises an amino acid residue, for example, as an amino acid component (AA)b' or (AA)b'', or as a hydrophobic component (X^{o'}), and wherein z' and j'' are 0, it is understood that the carbonyl adjacent to the caron defined by j'' is absent such that the N-terminal nitrogen of the amino acid (or N-terminal amino acid residue if more than one residue is present) is the nitrogen of the amide shown to be connected to (PEG)z'. Stated differently, it is understood that when z' is 0 and j'' is 0, the most N-terminal amino acid of L² forms and amide bond with the amino acid residue having the side chain comprising the cCPP.



[0178] In embodiments, M comprises -OH, , wherein y'' is an integer from 1-4. y'' can be 1. y'' can be 2. y'' can be 3. y'' can be 4.

[0179] x' can be an integer from 0 to 12. x' can be an integer from 1 to 12. x' can be an integer from 2 to 12. x' can be 0, 2, 4, 8, or 12. x' can be 0 or 2. x' can be 0, 2, or 12. x' can be 0 or 12. x' can be 2 or 12. x' can be 0. x' can be 1. x' can be 2. x' can be 3. x' can be 4. x' can be 5. x' can be 6. x' can be 7. x' can be 8. x' can be 9. x' can be 10. x' can be 11. x' can be 12.

[0180] j' can be 0, 1, or 2. j' can be 1. j' can be 2. It is understood that j' is 0 when x' is 0.

[0181] $\wedge$ can indicate L-stereochemistry. $\wedge$ can indicate D-stereochemistry.

[0182] y' can be an integer from 1 to 5, e.g., 1, 2, 3, 4, or 5, inclusive of all ranges and subranges therebetween. y' can be an integer from 2 to 5. y' can be an integer from 3 to 5. y' can be 3 or 4. y' can be 4 or 5. y' can be 1. y' can be 2. y' can be 3. y' can be 4. y' can be 5.

[0183] z' can be an integer from 0 to 12. z' can be an integer from 1 to 12. z' can be an integer from 2 to 12. z' can be 0, 2, 4, 8, or 12. z' can be 2 or 12. z' can be 0, 2, or 12. z' can be 0 or 12. z' can be 2 or 12. z' can be 0. z' can be 1. z' can be 2. z' can be 3. z' can be 4. z' can be 5. z' can be 6. z' can be 7. z' can be 8. z' can be 9. z' can be 10. z' can be 11. z' can be 12.

[0184] j'' can be 0, 1, or 2. j'' can be 0. j'' can be 1. j'' can be 2. It is understood that when z' is 0, j'' is 0.

[0185] z'' can be an integer from 0 to 12. z'' can be an integer from 1 to 12. z'' can be an integer from 2 to 12. z'' can be 0, 2, 4, 8, or 12. z'' can be 2 or 12. z'' can be 0, 2, or 12. z'' can be 0 or 12. z'' can be 2 or 12. z'' can be 0. z'' can be 1. z'' can be 2. z'' can be 3. z'' can be 4. z'' can be 5. z'' can be 6. z'' can be 7. z'' can be 8. z'' can be 9. z'' can be 10. z'' can be 11. z'' can be 12.

[0186] j''' can be 0, 1, or 2. j''' can be 0. j''' can be 1. j''' can be 2. It is understood that when z'' is 0, j''' is 0.

[0187] $K^\#$ indicates an L-lysine residue. $K^\#$ indicates a D-lysine residue.

[0188] a' can be 0 to 12. a' can be 1 to 12. a' can be 1 to 10. a' can be 1-8. a' can be 2 to 6. a' can be 2 to 4. a' can be 1. a' can be 2. a' can be 3. a' can be 4. a' can be 5. a' can be 6. a' can be 7. a' can be 8. a' can be 9. a' can be 10. a' can be 11. a' can be 12.

[0189] b' can be 0 to 12. b' can be 1 to 12. b' can be 1 to 10. b' can be 1-8. b' can be 2 to 6. b' can be 2 to 4. b' can be 1. b' can be 2. b' can be 3. b' can be 4. b' can be 5. b' can be 6. b' can be 7. b' can be 8. b' can be 9. b' can be 10. b' can be 11. b' can be 12.

[0190] b'' can be 0 to 12. b'' can be 1 to 12. b'' can be 1 to 10. b'' can be 1 to 8. b'' can be 2 to 6. b'' can be 2 to 4. b'' can be 1. b'' can be 2. b'' can be 3. b'' can be 4. a'' can be 5. b'' can be 6. b'' can be 7. b'' can be 8. b'' can be 9. b'' can be 10. b'' can be 11. b'' can be 12.

[0191] In embodiments, $\wedge$ indicates L-stereochemistry. In embodiments, $\wedge$ indicates D-stereochemistry.

[0192] The amino acids in (AA)a', (AA)b', or (AA)b'', can have D- or L- stereochemistry. The amino acid residues of (AA)a', (AA)b', or (AA)b'', independently, may all be D-amino acids.

The amino acid residues of (AA)a', (AA)b', or (AA)b'', independently, may all be L-amino acids.

[0193] (AA)a' can comprise one or more amino acid residues selected from lysine (K), arginine

(R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof. (AA)_a can comprise one or more amino acid residues selected from lysine

(K), arginine (R), histidine (H), glycine (G), β -alanine (B), or combinations thereof. (AA)a' can comprise one proline (P). (AA)a' can comprise one valine (V). (AA)a' can comprise one glycine

(G). (AA)a' can comprise one or more lysine (K). (AA)a' can comprise one lysine (K). (AA)a'

can comprise two lysine (K). (AA)a' can comprise three lysine (K). (AA)a' can comprise four lysine (K). (AA)a' can comprise one or more arginine (R). (AA)a' can comprise one arginine (R).

(AA)a' can comprise two arginine (R). (AA)a' can comprise three arginine (R). (AA)a' can

comprise four arginine (R). (AA)a' can comprise five arginine (R). (AA)a' can comprise six

arginine (R). (AA)a' can comprise one or more histidine (H). (AA)a' can comprise one histidine

(H). (AA)a' can comprise two histidine (H). (AA)a' can comprise three histidine (H). (AA)a' can

comprise four histidine (H). (AA)a' can comprise five histidine (H). (AA)a' can comprise six

histidine (H). (AA)a' can comprise one or more β -alanine (B). (AA)a' can comprise two β -

alanine (B). (AA)a' can comprise three β -alanine (B).

[0194] (AA)a' can comprise KK, KR, RR, RK, RF, HH, HK, HR, RH, KKK, KFK, KGK, KBK,

KBR, KRK, KRR, RKK, RRR, RHR, RRV, FRR, RFR, RBR, KKH, KHK, HKK, HRR, HRH,

HHR, HBH, HHH, HHHH, PKKK, KHKK, KKHK, KKKH, KHKH, HKHK, KKKK, KKRK,

KRKK, KRRK, KRKF, RKKR, RRRR, RBRB, BRBR, HRHR, RHRH, HBHB, BHBH, KGKK,

KKGK, HBHBH, RBHBH, BRBRB, HBRBH, BHRHR, BRHRH, HBKBH, RRRRR, KKKKK,

KKKRK, RKKKK, KRKKK, KKRKK, KKKKR, KKFRK, KBKKB, KRKIL, RBFBR,

RBRBR, RBHBR, RFRRF, RIRRI, RKKKKG, KRKKKG, KKRKKG, KKKKRG, KKKRKG,

RKKKKB, KRKKKB, KKRKKB, KKKKRB, KKKRKV, RRRRRR, HHHHHH, RHRHRH,

HRHRHR, KRKKKP, PKKKRK, KRKRK, RKRKR, RBRBRB, RBRRBR, KBKKB,

PKKKRKV, PGKKRKV, PKGKRKV, PKKGRKV, PKKKGKV, PKKKRGV or PKKKRKG.

(AA)a' can comprise R, RK, RF, KRK, KFK, KKRK, KKKR, or KKKRK. (AA)a' can comprise

RH, RHRH, or HRHR. (AA)a' can comprise HBHB, BHBH. (AA)a' can comprise RB, RBRB,

BRBR, BRBRB, RBRBR, RBRRBR. (AA)a' can comprise HBH, HBHBH, or HBRBH. (AA)a'

can comprise HHRBFBR. (AA)a' can comprise KBK, KBKBK or KBR. (AA)a' can comprise KGK or KGKK. (AA)a' can comprise KKGK, KKKK, KKKKR, KKKRK, KKKRKG, or KKRK. (AA)a' can comprise KR, KRK, or KRKKK. (AA)a' can comprise PGKKRKV. (AA)a' can comprise PKGKRKV, PKK, PKKGRKV, PKKK, PKKKGKV, PKKKRGV, PKKKRKG, PKKRGV, PKKRKG, or PKKRKV. (AA)a' can comprise RBHBH, RBHBR, RBR, RBRBR, or RBRBR. (AA)a' can comprise RFR. (AA)a' can comprise RHR. (AA)a' can comprise RKKK. (AA)a' can comprise RR, RRR, or RRRRFFF. In embodiments, (AA)a' can comprise RFRRF or RIRRI. In embodiments, (AA)a' can comprise KKFRK or KRKIL.

[0195] In embodiments, (AA)a' is an amino acid component comprising at least one amino acid residue. In embodiments, (AA)a' is an amino acid component comprising at least one amino acid residue with a charged side chain. In embodiments, (AA)a' is an amino acid component comprising at least one amino acid residue with a hydrophobic side chain. In embodiments, (AA)a' is an amino acid component comprising at least one amino acid residue with a charged side chain and at least one amino acid residue with a hydrophobic side chain. In embodiments, (AA)a' is an amino acid component comprising glycine (G), phenylalanine (F), β -alanine (B), arginine (R), glutamic acid (E) or combinations thereof. In embodiments, (AA)a' is an amino acid component comprising at least one glycine (G) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising at least one phenylalanine (F) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising one phenylalanine (F) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising two phenylalanine (F) amino acid residues. In embodiments, (AA)a' is an amino acid component comprising three phenylalanine (F) amino acid residues. In embodiments, (AA)a' is an amino acid component comprising at least one β -alanine (B) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising at least one arginine (R) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising one arginine (R) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising two arginine (R) amino acid residues. In embodiments, (AA)a' is an amino acid component comprising three arginine (R) amino acid residues. In embodiments, (AA)a' is an amino acid component comprising at least one arginine and at least one phenylalanine amino acid residue. In embodiments, (AA)a' is an amino acid component comprising one arginine and one phenylalanine amino acid residue. In embodiments, (AA)a' is an amino acid component comprising at least one arginine and at least

one glycine amino acid residue. In embodiments, (AA)a' is an amino acid component comprising at least two arginine and at least two glycine amino acid residues. In embodiments, (AA)a' is an amino acid component comprising at least three arginine and at least three glycine amino acid residues. In embodiments, (AA)a' is an amino acid component comprising at least four arginine and at least four glycine amino acid residues. In embodiments, (AA)a' comprises R, F, RF, RRF, RFR, GRF, BRF, RGRRG, or RFRRF. In embodiments, (AA)a' comprises GGRRGRRG or RFQILYBRBRB. In embodiments, (AA)a' comprises F. In embodiments, (AA)a' comprises F. In embodiments, (AA)a' comprises RF. In embodiments, (AA)a' comprises RRF. In embodiments, (AA)a' comprises RFR. In embodiments, (AA)a' comprises GRF. In embodiments, (AA)a' comprises BRF. In embodiments, (AA)a' comprises RGRRG. In embodiments, (AA)a' comprises RFRRF. In embodiments, (AA)a' comprises GGE. In embodiments, (AA)a' comprises GGRRGRRG. In embodiments, (AA)a' comprises RFQILYBRBRB.

[0196] (AA)b' can comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof. (AA)b' can comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), or combinations thereof. (AA)b' can comprise one proline (P). (AA)b' can comprise one valine (V). (AA)b' can comprise one glycine (G). (AA)b' can comprise one or more lysine (K). (AA)b' can comprise one lysine (K). (AA)b' can comprise two lysine (K). (AA)b' can comprise three lysine (K). (AA)b' can comprise four lysine (K). (AA)b' can comprise one or more arginine (R). (AA)b' can comprise one arginine (R). (AA)b' can comprise two arginine (R). (AA)b' can comprise three arginine (R). (AA)b' can comprise four arginine (R). (AA)b' can comprise five arginine (R). (AA)b' can comprise six arginine (R). (AA)b' can comprise one or more histidine (H). (AA)b' can comprise one histidine (H). (AA)b' can comprise two histidine (H). (AA)b' can comprise three histidine (H). (AA)b' can comprise four histidine (H). (AA)b' can comprise five histidine (H). (AA)b' can comprise six histidine (H). (AA)b' can comprise one or more β -alanine (B). (AA)b' can comprise two β -alanine (B). (AA)b' can comprise three β -alanine (B).

[0197] (AA)b' can comprise KK, KR, RR, RK, RF, HH, HK, HR, RH, KKK, KFK, KGK, KBK, KBR, KRK, KRR, RKK, RRR, RHR, RRV, FRR, RFR, RBR, KKH, KHK, HKK, HRR, HRH, HHR, HBH, HHH, HHHH, PKKK, KHKK, KKKH, KHKH, HKHK, KKKK, KKRK, KRKK, KRRK, KRKF, RKKR, RRRR, RBRB, BRBR, HRHR, RHRH, HBHB, BHBH, KGKK,

KKGK, HBHBH, RBHBH, BRBRB, HBRBH, BHRHR, BRHRH, HBKBH, RRRRR, KKKKK, KKKRK, RKKKK, KRKKK, KKRKK, KKKKR, KKFRK, KBKKB, KRKIL, RBFBR, RBRBR, RBHBR, RFRRF, RIRRI, RKKKKG, KRKKKG, KKRKKG, KKKKRG, KKKRKG, RKKKKB, KRKKKB, KKRKKB, KKKKRB, KKKRKV, RRRRRR, HHHHHH, RHRHRH, HRHRHR, KRKKKP, PKKKRK, KRKRKR, RKRKRK, RBRBRB, RRRRBR, KBKKBK, PKKKRKV, PGKKRKV, PKGKRKV, PKKGRKV, PKKKGKV, PKKKRGV or PKKKRKG. (AA)b' can comprise R, RK, RF, KRK, KFK, KKRK, KKKR, or KKKRK. (AA)b' can comprise RH, RHRH, or HRHR. (AA)b' can comprise HBHB, BHBH. (AA)b' can comprise RB, RBRB, BRBR, BRBRB, RBRBR, RRRRBR. (AA)b' can comprise HBH, HBHBH, or HBRBH. (AA)b' can comprise HHRBFBR. (AA)b' can comprise KBK, KBKKB or KBR. (AA)b' can comprise KGK or KGKK. (AA)b' can comprise KKGK, KKKK, KKKKR, KKKRK, KKKRKG, or KKRK. (AA)b' can comprise KR, KRK, or KRKKK. (AA)b' can comprise PGKKRKV. (AA)b' can comprise PKGKRKV, PKK, PKKGRKV, PKKK, PKKKGKV, PKKKRGV, PKKKRKG, PKKRGV, PKKRKG, or PKKRKV. (AA)b' can comprise RBHBH, RBHBR, RBR, RBRBR, or RRRRBR. (AA)b' can comprise RFR. (AA)b can comprise RHR. (AA)b' can comprise RKKK. (AA)b' can comprise RR, RRR, or RRRRFFF. In embodiments, (AA)b' can comprise RFRRF or RIRRI. In embodiments, (AA)b' can comprise KKFRK or KRKIL.

[0198] In embodiments, (AA)b' is an amino acid component comprising at least one amino acid residue. In embodiments, (AA)b' is an amino acid component comprising at least one amino acid residue with a charged side chain. In embodiments, (AA)b' is an amino acid component comprising at least one amino acid residue with a hydrophobic side chain. In embodiments, (AA)b' is an amino acid component comprising at least one amino acid residue with a charged side chain and at least one amino acid residue with a hydrophobic side chain. In embodiments, (AA)b' is an amino acid component comprising glycine (G), phenylalanine (F), β -alanine (B), arginine (R), glutamic acid (E) or combinations thereof. In embodiments, (AA)b' is an amino acid component comprising at least one glycine (G) amino acid residue. In embodiments, (AA)b' is an amino acid component comprising at least one phenylalanine (F) amino acid residue. In embodiments, (AA)b' is an amino acid component comprising one phenylalanine (F) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising two phenylalanine (F) amino acid residues. In embodiments, (AA)a' is an amino acid component comprising three phenylalanine (F) amino acid residues. In embodiments, (AA)a is an amino acid component

comprising at least one β -alanine (B) amino acid residue. In embodiments, (AA)b' is an amino acid component comprising at least one arginine (R) amino acid residue. In embodiments, (AA)b' is an amino acid component comprising at least one arginine (R) amino acid residue. In embodiments, (AA)b' is an amino acid component comprising two arginine (R) amino acid residues. In embodiments, (AA)b' is an amino acid component comprising three arginine (R) amino acid residues. In embodiments, (AA)b' is an amino acid component comprising at least one arginine and at least one phenylalanine amino acid residue. In embodiments, (AA)b' is an amino acid component comprising one arginine and one phenylalanine amino acid residue. In embodiments, (AA)b' is an amino acid component comprising at least one arginine and at least one glycine amino acid residue. In embodiments, (AA)b' is an amino acid component comprising at least two arginine and at least two glycine amino acid residues. In embodiments, (AA)b' is an amino acid component comprising at least three arginine and at least three glycine amino acid residues. In embodiments, (AA)b' is an amino acid component comprising at least four arginine and at least four glycine amino acid residues. In embodiments, (AA)b' comprises R, F, RF, RRF, RFR, GRF, BRf, RGRRG, or RFRRF. In embodiments, (AA)b' comprises GGRRGRRG or RFQILYBRBRB. In embodiments, (AA)b' comprises F. In embodiments, (AA)b' comprises F. In embodiments, (AA)b' comprises RF. In embodiments, (AA)b' comprises RRF. In embodiments, (AA)b' comprises RFR. In embodiments, (AA)b' comprises GRF. In embodiments, (AA)b' comprises BRf. In embodiments, (AA)b' comprises RGRRG. In embodiments, (AA)b' comprises RFRRF. In embodiments, (AA)b' comprises GGE. In embodiments, (AA)b' comprises GGRRGRRG. In embodiments, (AA)b' comprises RFQILYBRBRB.

[0199] (AA)b'' can comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof. (AA)b'' can comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), or combinations thereof. (AA)b'' can comprise one proline (P). (AA)b'' can comprise one valine (V). (AA)b'' can comprise one glycine (G). (AA)b'' can comprise one or more lysine (K). (AA)b'' can comprise one lysine (K). (AA)b'' can comprise two lysine (K). (AA)b'' can comprise three lysine (K). (AA)b'' can comprise four lysine (K). (AA)b'' can comprise one or more arginine (R). (AA)b'' can comprise one arginine (R). (AA)b'' can comprise two arginine (R). (AA)b'' can comprise three arginine

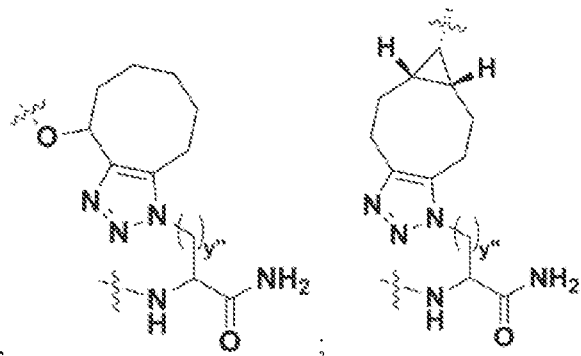
(R). (AA)b'' can comprise four arginine (R). (AA)b'' can comprise five arginine (R). (AA)b'' can comprise six arginine (R). (AA)b'' can comprise one or more histidine (H). (AA)b'' can comprise one histidine (H). (AA)b'' can comprise two histidine (H). (AA)b'' can comprise three histidine (H). (AA)b'' can comprise four histidine (H). (AA)b'' can comprise five histidine (H). (AA)b'' can comprise six histidine (H). (AA)b'' can comprise one or more β -alanine (B). (AA)b'' can comprise two β -alanine (B). (AA)b'' can comprise three β -alanine (B).

[0200] (AA)b'' can comprise KK, KR, RR, RK, RF, HH, HK, HR, RH, KKK, KFK, KGK, KBK, KBR, KRK, KRR, RKK, RRR, RHR, RRV, FRR, RFR, RBR, KKH, KHK, HKK, HRR, HRH, HHR, HBH, HHH, HHHH, PKKK, KHKK, KKHK, KKKH, KHKH, HKHK, KKKK, KKRK, KRKK, KRRK, KRKF, RKKR, RRRR, RBRB, BRBR, HRHR, RHRH, HBHB, BHBH, KGKK, KKGK, HBHBH, RBHBH, BRBRB, HBRBH, BHRHR, BRHRH, HBKBH, RRRRR, KKKKK, KKKRK, RKKKK, KRKKK, KKRKK, KKKKR, KKFRK, KBKKB, KRKIL, RBFBR, RBRBR, RBHBR, RFRRF, RIRRI, RKKKKG, KRKKKG, KKRKKG, KKKKRG, KKKRKG, RKKKKB, KRKKKB, KKRKKB, KKKKRB, KKKRKV, RRRRRR, HHHHHH, RHRHRH, HRHRHR, KRKKKP, PKKKRK, KRKRK, RKRK, RBRBRB, RBRBR, KBKKB, PKKKRKV, PGKKRKV, PKGKRKV, PKKGRKV, PKKKGKV, PKKKRGV or PKKKRKG. (AA)b'' can comprise R, RK, KRK, KKRK, KKKR, or KKKRK. (AA)b'' can comprise RH, RHRH, or HRHR. (AA)b'' can comprise HBHB, BHBH. (AA)b'' can comprise RB, RBRB, BRBR, BRBRB, RBRBR, RBRRBR. (AA)b'' can comprise HBH, HBHBH, or HBRBH. (AA)b'' can comprise HHRBFBR. (AA)b'' can comprise KBK, KBKKB or KBR. (AA)b'' can comprise KGK or KGKK. (AA)b'' can comprise KKGK, KKKK, KKKKR, KKKRK, KKKRKG, or KKRK. (AA)b'' can comprise KR, KRK, or KRKKK. (AA)b'' can comprise PGKKRKV. (AA)b'' can comprise PKGKRKV, PKK, PKKGRKV, PKKK, PKKKGKV, PKKKRGV, PKKKRKG, PKKRGV, PKKRKG, or PKKRKV. ((AA)b'' can comprise RBHBH, RBHBR, RBR, RBRBR, or RBRRBR. (AA)b'' can comprise RFR. (AA)b'' can comprise RHR. (AA)b'' can comprise RKKK. (AA)b'' can comprise RR, RRR, or RRRRFFF. In embodiments, (AA)b'' can comprise RFRRF or RIRRI. In embodiments, (AA)b'' can comprise KKFRK or KRKIL.

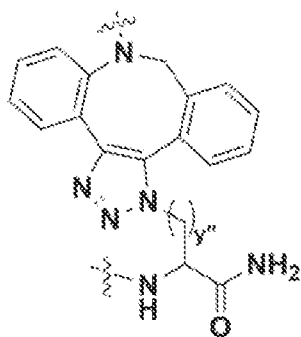
[0201] In embodiments, (AA)b'' is an amino acid component comprising at least one amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising at least one amino acid residue with a charged side chain. In embodiments, (AA)b'' is an amino acid component comprising at least one amino acid residue with a hydrophobic side chain. In embodiments,

(AA)b'' is an amino acid component comprising at least one amino acid residue with a charged side chain and at least one amino acid residue with a hydrophobic side chain. In embodiments, (AA)b'' is an amino acid component comprising glycine (G), phenylalanine (F), β -alanine (B), arginine (R), glutamic acid (E) or combinations thereof. In embodiments, (AA)b'' is an amino acid component comprising at least one glycine (G) amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising at least one phenylalanine (F) amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising one phenylalanine (F) amino acid residue. In embodiments, (AA)a' is an amino acid component comprising two phenylalanine (F) amino acid residues. In embodiments, (AA)a' is an amino acid component comprising three phenylalanine (F) amino acid residues. In embodiments, (AA) α is an amino acid component comprising at least one β -alanine (B) amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising at least one arginine (R) amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising at least one arginine (R) amino acid residue. In embodiments, (AA)b' is an amino acid component comprising two arginine (R) amino acid residues. In embodiments, (AA)b' is an amino acid component comprising three arginine (R) amino acid residues. In embodiments, (AA)b' is an amino acid component comprising at least one arginine and at least one phenylalanine amino acid residue. In embodiments, (AA)b' is an amino acid component comprising one arginine and one phenylalanine amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising at least one arginine and at least one glycine amino acid residue. In embodiments, (AA)b'' is an amino acid component comprising at least two arginine and at least two glycine amino acid residues. In embodiments, (AA)b'' is an amino acid component comprising at least three arginine and at least three glycine amino acid residues. In embodiments, (AA)b'' is an amino acid component comprising at least four arginine and at least four glycine amino acid residues. In embodiments, (AA)b'' comprises R, F, RF, RRF, RFR, GRF, BRF, RGRRG, or RFRRF. In embodiments, (AA)b'' comprises GGRRGRRG or RFQILYBRBRB. In embodiments, (AA)b'' comprises F. In embodiments, (AA)b'' comprises F. In embodiments, (AA)b'' comprises RF. In embodiments, (AA)b'' comprises RRF. In embodiments, (AA)b'' comprises RFR. In embodiments, (AA)b'' comprises GRF. In embodiments, (AA)b'' comprises BRF. In embodiments, (AA)b'' comprises RGRRG. In embodiments, (AA)b'' comprises RFRRF.

In embodiments, (AA)b'' comprises GGE. In embodiments, (AA)b'' comprises GGRRGRRG. In embodiments, (AA)b'' comprises RFQILYBRBRB.



[0202] In embodiments, M' comprises -NH-, -O-,



or , wherein y'' is an integer from 1 to 4. AAsc can be a side chain or terminus of an amino acid residue on the cCPP. Non-limiting examples of AAsc include aspartic acid, glutamic acid, glutamine, asparagine, or lysine, or a modified side chain of glutamine or asparagine (e.g., a reduced side chain having an amino group). AAsc can be an AAsc as defined herein.

[0203] y'' can be an integer from 0 to 10, e.g., 0, 1, 2, 3, 4, 5, 6, 7, 8, 9, and 10. y'' can be 0, 1, 2, 3, or 4. y'' can be 0. y'' can be 1. y'' can be 2. y'' can be 3. y'' can be 4.

[0204] X^{o'} can be an aliphatic hydrocarbon. X^{o'} can be a C₄-C₁₂ aliphatic hydrocarbon. X^{o'} can be a C₄-C₈ aliphatic hydrocarbon. X^{o'} can be a C₆ aliphatic hydrocarbon. X^{o'} can be selected from phenylalanine, 3-(4',4'-biphenyl)-L-alanine, tryptophan, tyrosine, valine, isoleucine, leucine, or histidine, or any combinations thereof. X^{o'} can be amino acid residue selected from tryptophan, tyrosine, isoleucine, leucine, histidine, phenylalanine, or any combinations thereof.

[0205] X^{o'} can be a D- or L-amino acid residue with a hydrophobic side chain. X^{o'} can be a naturally occurring or a non-naturally occurring amino acid residue with a hydrophobic side chain. X^{o'} can be an amino acid residue with an aromatic side chain. X^{o'} can be an amino acid residue with a heteroaromatic side chain. X^{o'} can be selected from phenylalanine; 2-naphthylalanine (Nal

or nal); 3-(4',4-biphenyl)-L-alanine (Bip or bip); tryptophan; tyrosine; valine; isoleucine; leucine; or histidine, or any combinations thereof. X^{o'} can be 2-naphtylalanine (Nal or nal). X^{o'} can be L-2-naphtylalanine (L-Nal or Nal). X^{o'} can be D-2-naphtylalanine (d-nal or nal). X^{o'} can be 3-(4',4-biphenyl)-L-alanine. X^{o'} can be 3-(4',4-biphenyl)-L-alanine (L-Bip or Bip). X^{o'} can be 3-(4',4-biphenyl)-D-alanine (d-bip or bip). X^{o'} can be a C₄-C₈ alkyl or dialkyl hydrocarbon. X^{o'} can be a C₆ alkyl or dialkyl hydrocarbon.

[0206] X^{o''} can be an aliphatic hydrocarbon. X^{o''} can be a C₄-C₁₂ aliphatic hydrocarbon. X^{o''} can be a C₄-C₈ aliphatic hydrocarbon. X^{o''} can be a C₆ aliphatic hydrocarbon. X^{o''} can be selected from phenylalanine, 3-(4',4-biphenyl)-L-alanine, tryptophan, tyrosine, valine, isoleucine, leucine, or histidine, or any combinations thereof. X^{o''} can be amino acid residue selected from tryptophan, tyrosine, isoleucine, leucine, histidine, phenylalanine, or any combinations thereof.

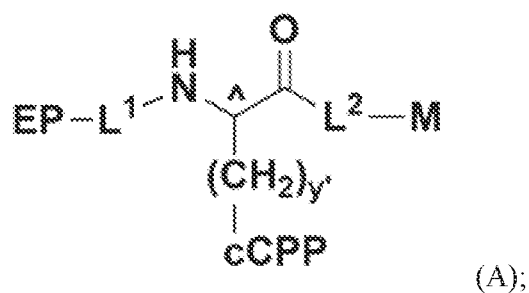
[0207] X^{o'''} can be a D- or L-amino acid residue with a hydrophobic side chain. X^{o'''} can be a naturally occurring or a non-naturally occurring amino acid residue with a hydrophobic side chain. X^{o'''} can be an amino acid residue with an aromatic side chain. X^{o'''} can be an amino acid residue with a heteroaromatic side chain. X^{o'''} can be selected from phenylalanine; 2-naphtylalanine (Nal or nal); 3-(4',4-biphenyl)-L-alanine (Bip or bip); tryptophan; tyrosine; valine; isoleucine; leucine; or histidine, or any combinations thereof. X^{o'''} can be 2-naphtylalanine (Nal or nal). X^{o'''} can be L-2-naphtylalanine (L-Nal or Nal). X^{o'''} can be D-2-naphtylalanine (d-nal or nal). X^{o'''} can be 3-(4',4-biphenyl)-L-alanine. X^{o'''} can be 3-(4',4-biphenyl)-L-alanine (L-Bip or Bip). X^{o'''} can be 3-(4',4-biphenyl)-D-alanine (dBip or bip). X^{o'''} can be a C₄-C₈ alkyl or dialkyl hydrocarbon. X^{o'''} can be a C₆ alkyl or dialkyl hydrocarbon.

Endosomal Escape Vehicles (EEVs)

[0208] EEVs comprising a cyclic cell penetrating peptide (cCPP) and one or more linkers are provided. In embodiments, the EEV includes an exocyclic peptide (EP). In embodiments, the EP is absent.

[0209] An EEV can comprise the structure of Formula (A):

Formula (A):



wherein:

EP is a linear exocyclic peptide;

5 cCPP is a cell penetrating peptide;

L¹ and L², are, independently, a linker arm;

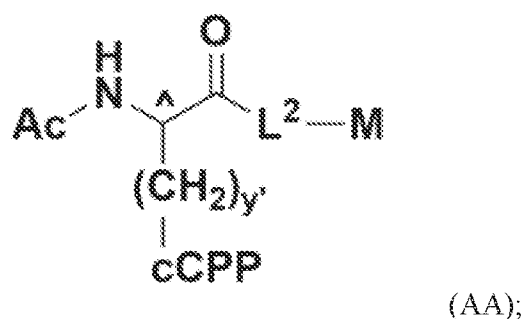
^ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5; and

M comprises a reactive handle.

10 [0210] An EEV can comprise the structure of Formula (AA):

Formula (AA):



wherein:

cCPP is a cell penetrating peptide;

15 Ac is acetyl;

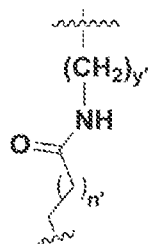
L² is a linker arm;

^ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5; and

M comprises a reactive handle.

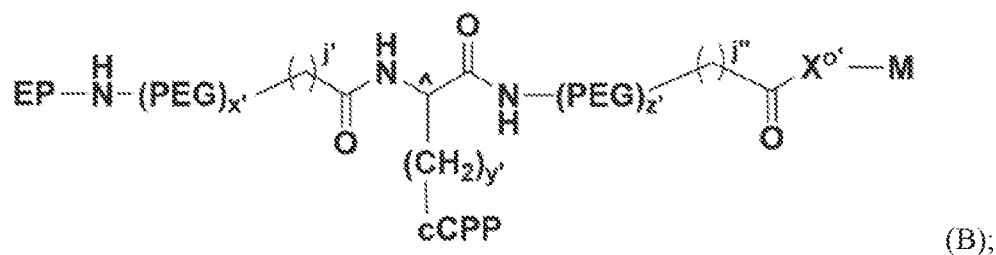
[0211] In embodiments, the cCPP is linked to the $-(CH_2)_{y'}$ - group through the AA_{SC} of the cCPP. In embodiments, the cCPP is linked to the $-(CH_2)_{y'}$ - group through the AA_{SC} of the cCPP such that



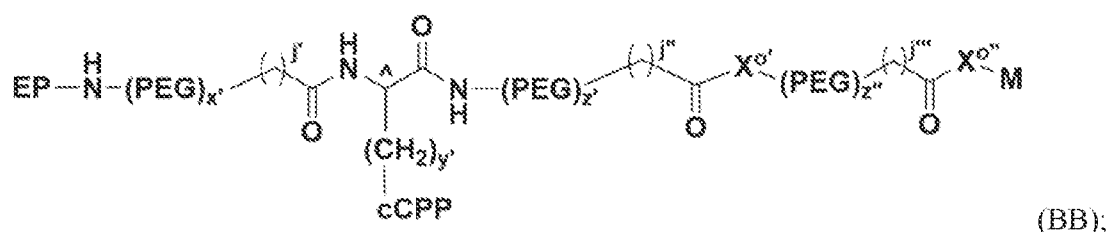
the connection is of the structure wherein n' is an integer from 1 to 5. In embodiments, n' is 1. In embodiments, n' is 2. In embodiments, n' is 3. In embodiment, n' is 4. In embodiments, n' is 5. In embodiments when n' is 1, the AA_{SC} is the side chain of a glutamine residue. In embodiments when the AA_{SC} is the side chain of glutamine, the carboxamide amide nitrogen of the glutamine side chain forms a bond with the $-(CH_2)_{y'}$ - group.

[0212] In embodiments, the EEV comprises a structure selected from:

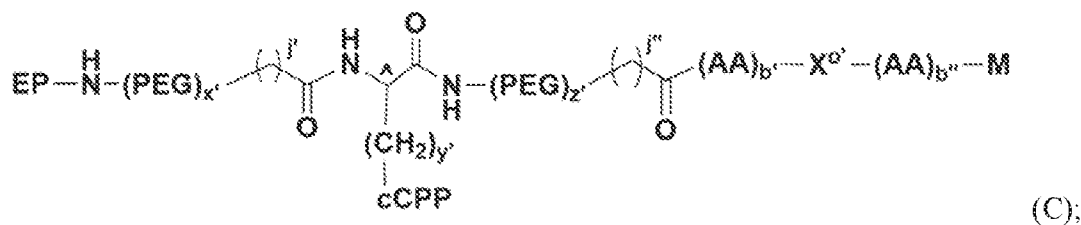
Formula (B):



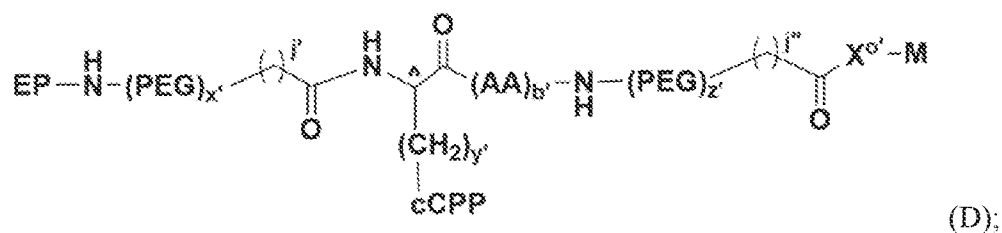
Formula (BB):



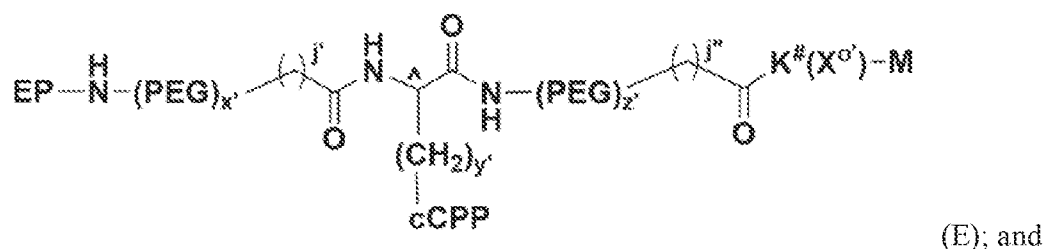
Formula (C):



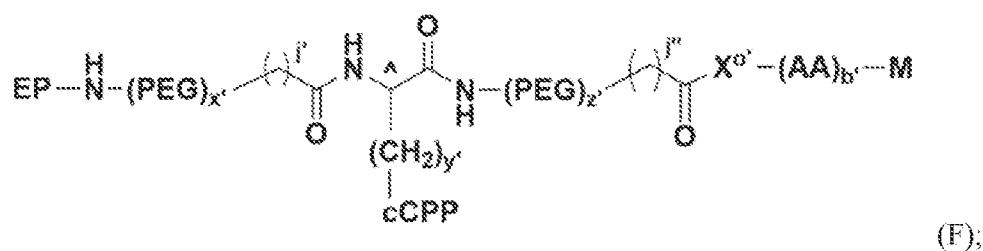
Formula (D):



Formula (E):



5 Formula (F):



wherein:

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

10 x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

$\wedge$ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5;

z' is an integer from 0 to 12;

15 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$\text{X}^{o'}$ and $\text{X}^{o''}$ each independently comprise a hydrophobic component;

$\text{K}^{\#}$ is D-lysine or L-lysine residue;

(AA) b' and (AA) b'' are each an amino acid (AA) component comprising at least one amino acid residue;

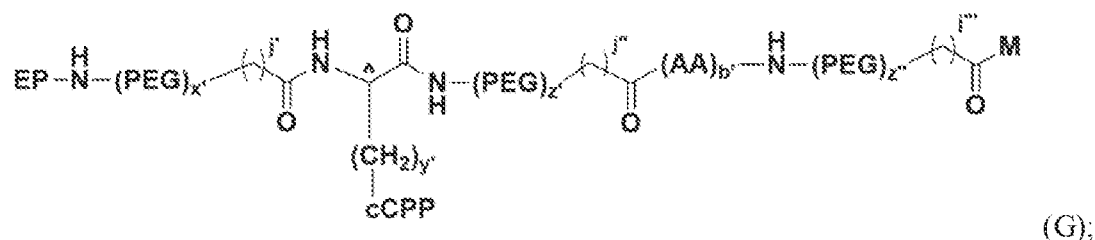
b' is an integer from 0 to 12;

b'' is an integer from 0 to 12; and

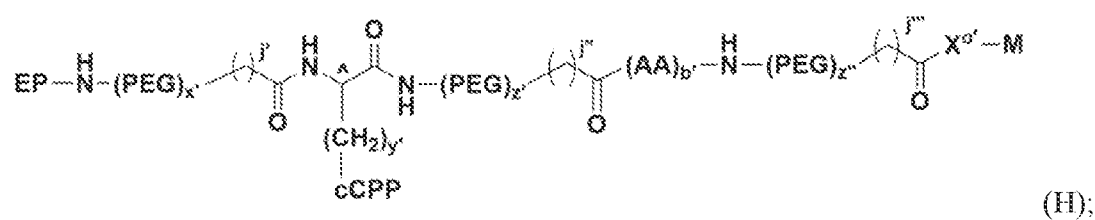
5 M comprises a reactive handle.

[0213] In embodiments, the EEV comprises a structure selected from:

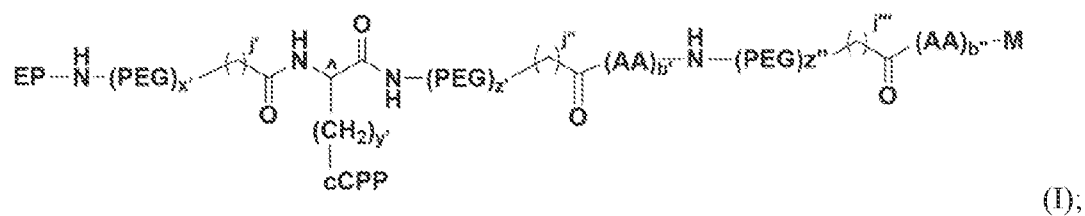
Formula (G):



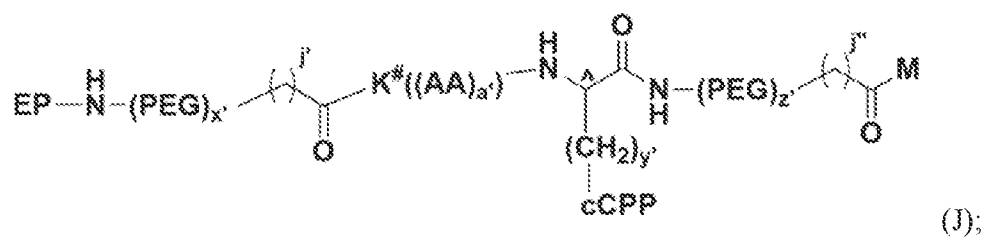
Formula (H):



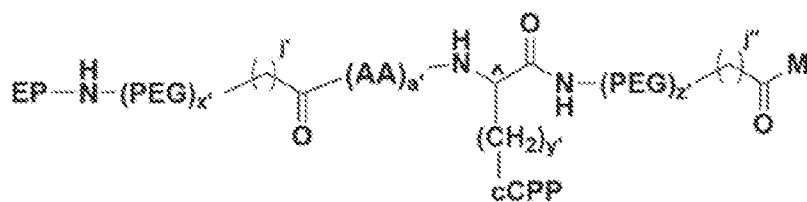
Formula (I):



Formula (J):

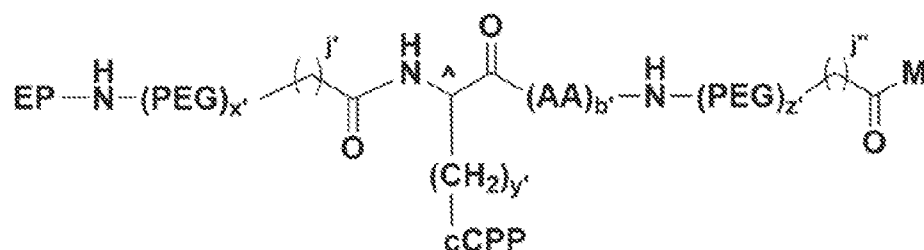


Formula (JJ):



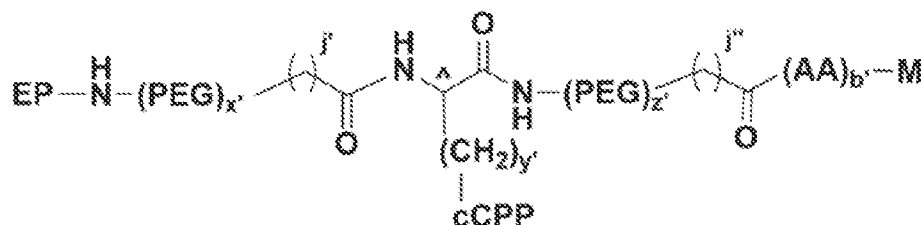
(JJ);

Formula (K):



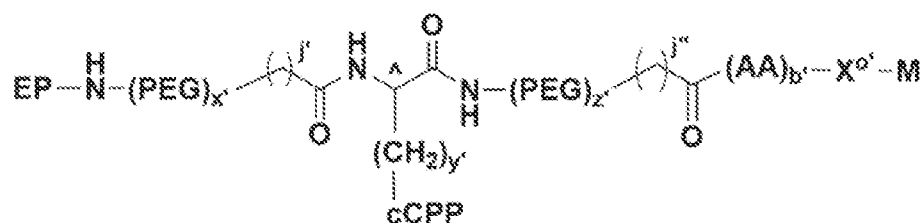
(K);

5 Formula (L):



(L);

Formula (M):



(M);

wherein:

- 10 EP is a linear exocyclic peptide;
- cCPP is a cyclic cell penetrating peptide;
- x' is an integer from 0 to 12;
- j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

^ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

5 z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

X^{o'} comprises a hydrophobic component;

K[#] is D-lysine or L-lysine residue;

(AA)a', (AA)b', and (AA)b'' are each an amino acid (AA) component comprising at least one

10 amino acid residue;

a' is an integer from 0 to 12;

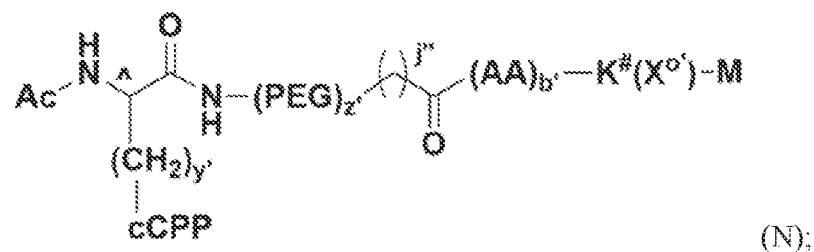
b' is an integer from 0 to 12;

b'' is an integer from 0 to 12; and

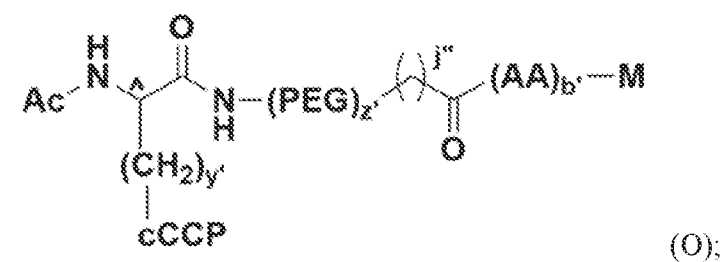
M comprises a reactive handle.

15 [0214] In embodiments, the EEV comprises a structure selected from:

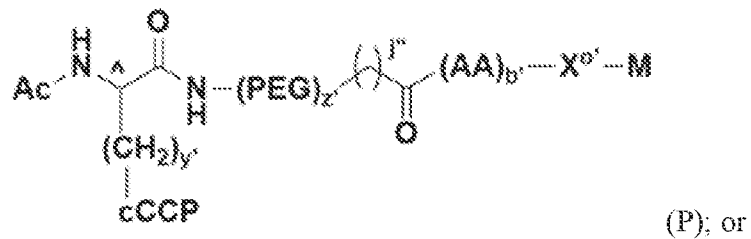
Formula (N):



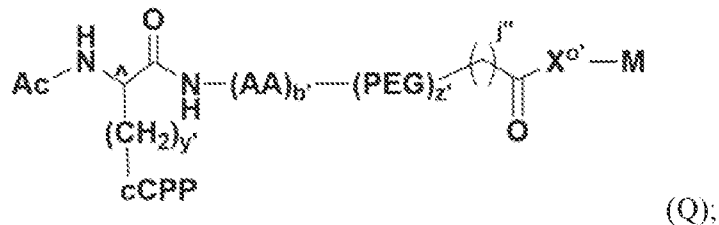
Formula (O):



Formula (P):



Formula (Q):



5 wherein:

Ac is acetyl;

cCPP is a cyclic cell penetrating peptide;

y' is an integer from 1 to 5;

^ indicates L- or D-stereochemistry;

10 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^{o'} is a hydrophobic component;

K[#] is D-lysine or L-lysine residue;

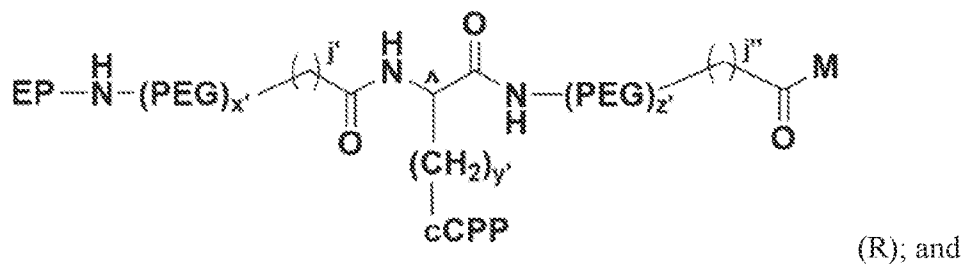
(AA)_{b'} is an amino acid (AA) component comprising at least one amino acid residue;

15 b' is an integer from 0 to 12; and

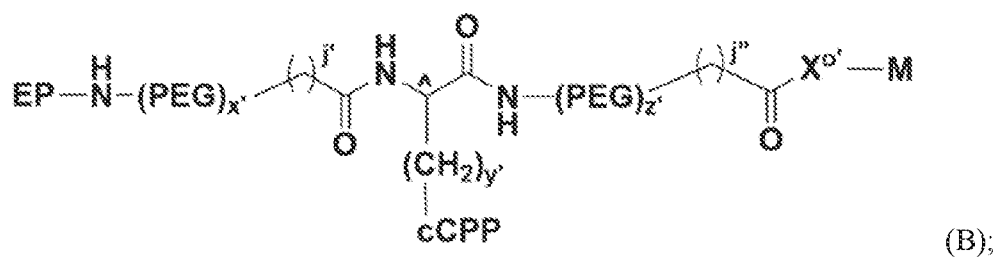
M comprises a reactive handle.

[0215] In embodiments, the EEV comprises a structure selected from Formula (R) and Formula (B):

Formula (R):



Formula (B):



5 wherein

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

10 $\wedge$ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5;

z' is an integer from 0 to 12;

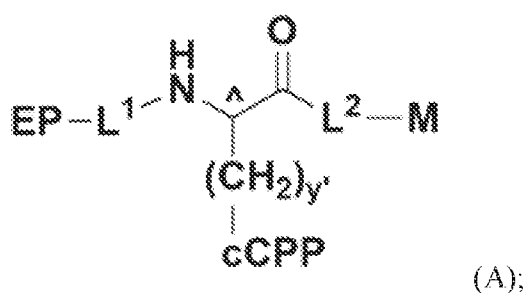
j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$\text{X}^{o'}$ comprises a hydrophobic component; and

15 M comprises a reactive handle.

[0216] An EEV can comprise the structure of Formula (A):

Formula (A):



wherein

EP is a linear exocyclic peptide;

cCPP is a cell penetrating peptide;

L^1 and L^2 , are, independently, a linker arm;

5 ^ indicates D-stereochemistry;

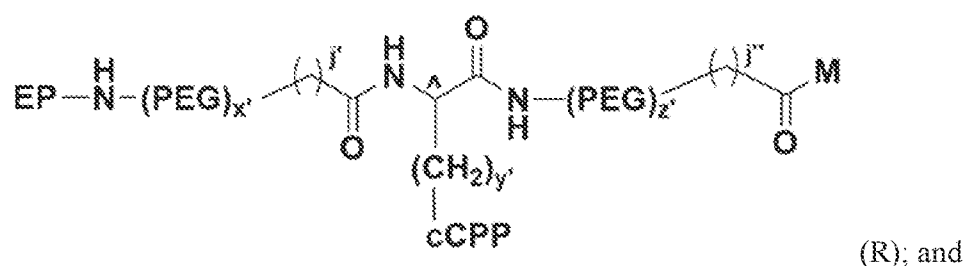
y' is an integer from 1 to 5;

M comprises a reactive handle; and

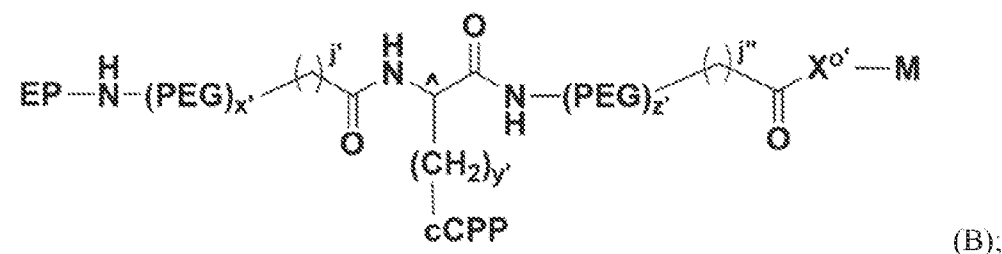
wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to
10 the linker.

[0217] In embodiments, the EEV of Formula (A) comprises a structure selected from Formula (R) and Formula (B):

Formula (R):



15 Formula (B):



wherein:

EP is a linear exocyclic peptide (EP);

cCPP a cell penetrating peptide (cCPP);

20 x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

^ indicates D-stereochemistry;

y' is an integer from 1 to 5;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$X^{o'}$ comprises a hydrophobic component; and

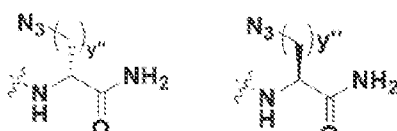
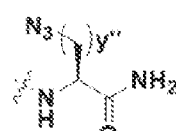
M comprises a reactive handle,

- 5 wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0218] X can be a D or L amino acid residue with a hydrophobic side chain. X can be a naturally occurring or a non-naturally occurring amino acid residue with a hydrophobic side chain. X can be an amino acid residue with an aromatic side chain. X can be an amino acid residue with a heteroaromatic side chain. X can be 2-naphtylalanine. X can be Nal. X can be d-Nal (nal). X can be 3-(4-biphenyl)-D-alanine. X can be Bip. X can be bip. X can be a C₄-C₈ alkyl hydrocarbon. X can be a C₆ alkyl hydrocarbon. X can be amino acid selected from tryptophan, tyrosine, isoleucine, leucine, histidine, phenylalanine, or a combination thereof.

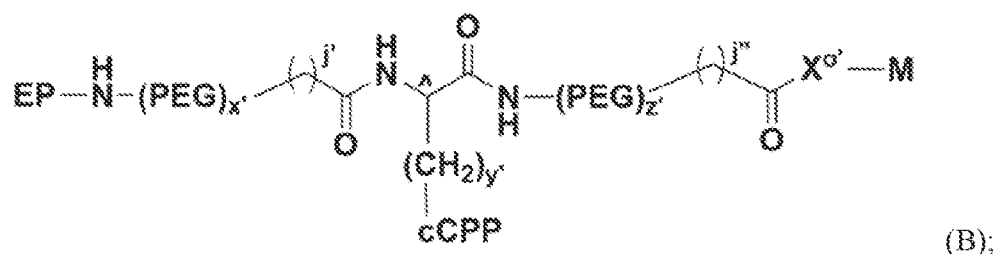
15 [0219] In embodiments, L¹ and L², when present, are each, independently, a hydrocarbon linker (e.g., NR_hH-(CH₂)_n-COOH), a PEG component (e.g., NR_hH-(CH₂O)_n-COOH, wherein R_h is H, methyl or ethyl), a linker comprising a hydrophobic component, one or more amino acid residue, or a combination thereof.

[0220] In embodiments, AAsc is a side chain of glutamine. In embodiments, AAsc is a side chain of L-glutamine. In embodiments, AAsc is a side chain of D-glutamine.

[0221] In embodiments, M comprises -OH or , or , wherein y'' is an integer from 1 to 4.

[0222] In embodiments, the EEV comprises Formula (B):

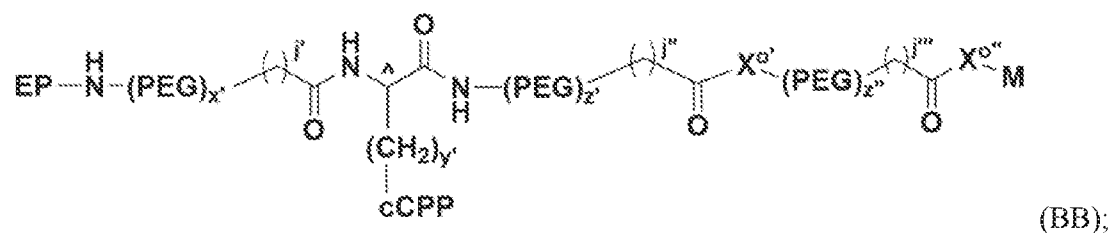
Formula (B):



wherein EP, cCPP, x' , j' , $\wedge$, y' , z' , j'' , $X^{o'}$, and M are as defined herein.

[0223] In embodiments, the EEV comprises Formula (BB):

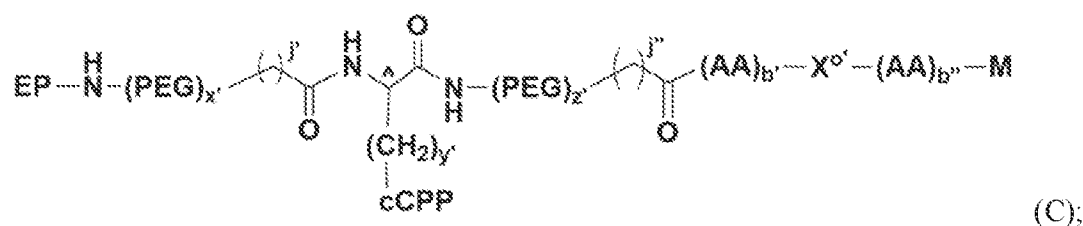
Formula (BB):



5 wherein EP, cCPP, x' , j' , $\wedge$, y' , z' , j'' , z'' , j''' , $X^{o'}$, $X^{o''}$, and M are as defined herein.

[0224] In embodiments, the EEV comprises Formula (C):

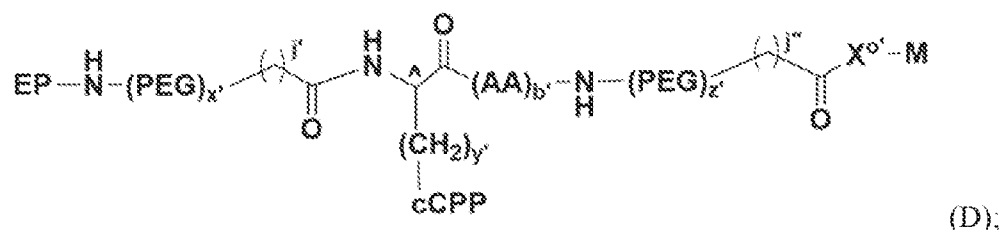
Formula (C):



wherein EP, cCPP, x' , j' , $\wedge$, y' , z' , j'' , $X^{o'}$, $(\text{AA})_{b'}$, $(\text{AA})_{b''}$, b' , b'' , and M are as defined herein.

10 [0225] In embodiments, the EEV comprises Formula (D):

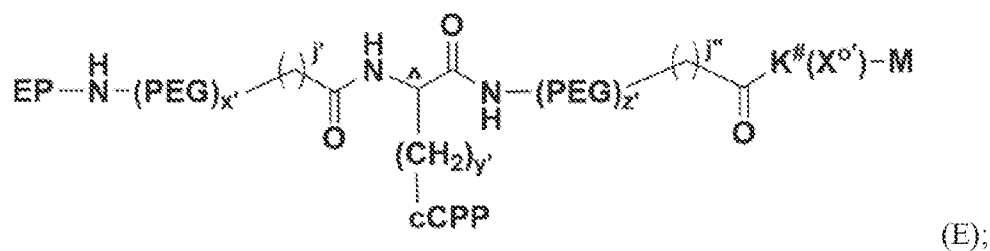
Formula (D):



wherein EP, cCPP, x' , j' , $\wedge$, y' , z' , j'' , $X^{o'}$, $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0226] In embodiments, the EEV comprises Formula (E):

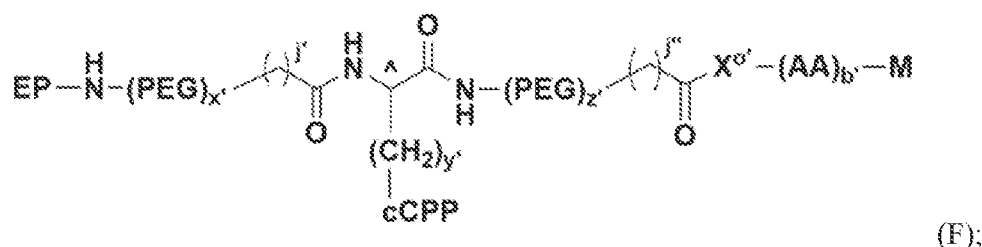
Formula (E):



wherein EP, cCPP, x' , j' , y' , z' , j'' , $\text{X}^{\sigma'}$, $\text{K}^{\#}$, and M are as defined herein.

[0227] In embodiments, the EEV comprises Formula (F):

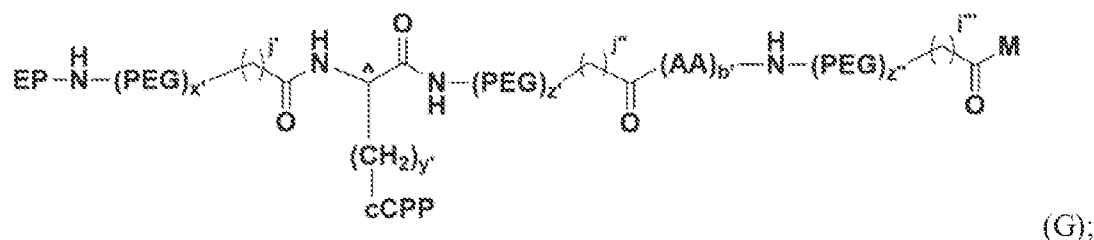
5 Formula (F):



wherein EP, cCPP, x' , j' , y' , z' , j'' , $\text{X}^{\sigma'}$, $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0228] In embodiments, the EEV comprises Formula (G):

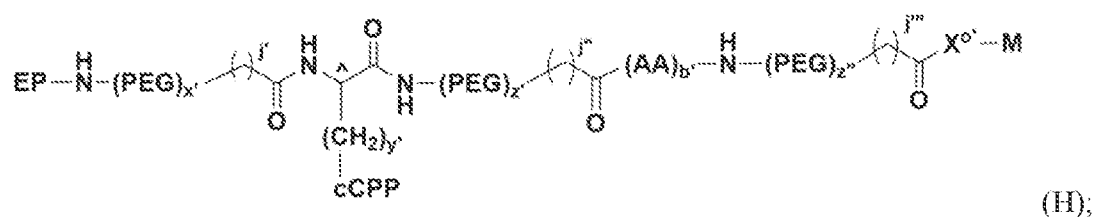
Formula (G):



wherein EP, cCPP, x' , j' , y' , z' , j'' , z'' , j''' , $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0229] In embodiments, the EEV comprises Formula (H):

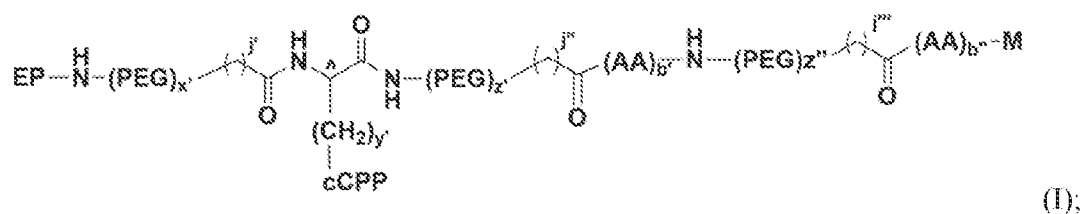
Formula (H):



wherein EP, cCPP, x' , j' , y' , z' , j'' , z'' , j''' , $\text{X}^{\sigma'}$, $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0230] In embodiments, the EEV comprises Formula (I):

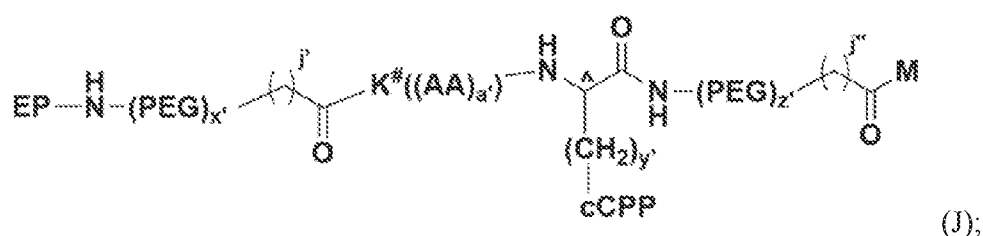
Formula (1):



wherein EP, cCPP, x' , j' , $\wedge$, y' , z' , j'' , z'' , j''' , (AA)b', (AA)b'', b', b'', and M are as defined herein.

[0231] In embodiments, the EEV comprises Formula (J):

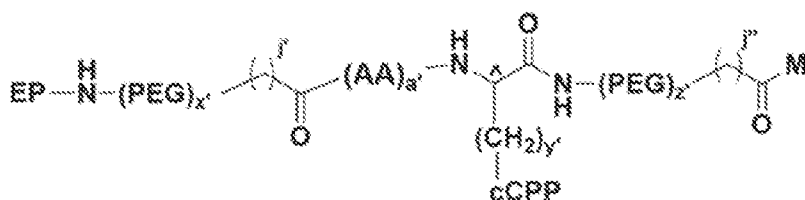
5 *Formula (J):*



wherein EP, cCPP, x' , j' , $K^\#$, $(AA)a'$, a' , $\wedge$, y' , z' , j'' , and M are as defined herein.

[0232] In embodiments, the EEV comprises Formula (JJ):

Formula (JJ):

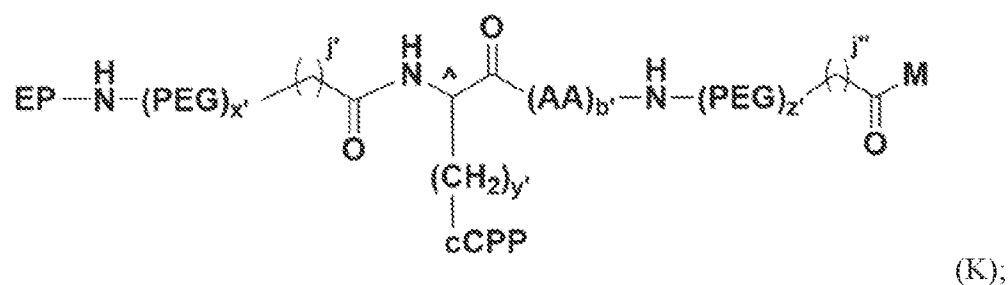


10 (II);

wherein EP, cCPP, x' , j' , $(AA)a'$, a' , $\wedge$, y' , z' , j'' , and M are as defined herein.

[0233] In embodiments, the EEV comprises Formula (K):

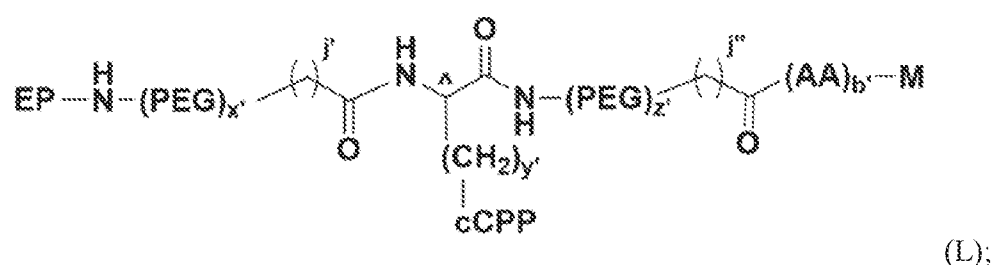
Formula (K):



wherein EP, cCPP, x' , j' , y' , z' , j'' , $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0234] In embodiments, the EEV comprises Formula (L):

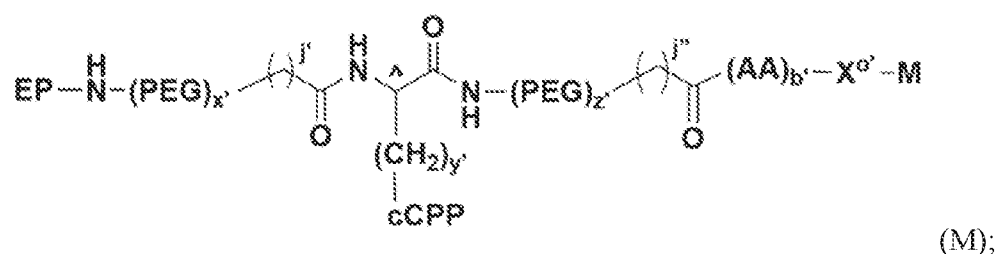
5 Formula (L):



wherein EP, cCPP, x' , j' , y' , z' , j'' , $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0235] In embodiments, the EEV comprises Formula (M):

Formula (M):

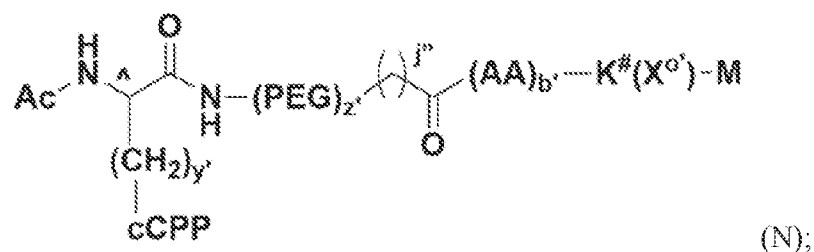


10

wherein EP, cCPP, x' , j' , y' , z' , j'' , $\text{X}^{o'}$, $(\text{AA})_{b'}$, b' , and M are as defined herein.

[0236] In embodiments, the EEV comprises Formula (N):

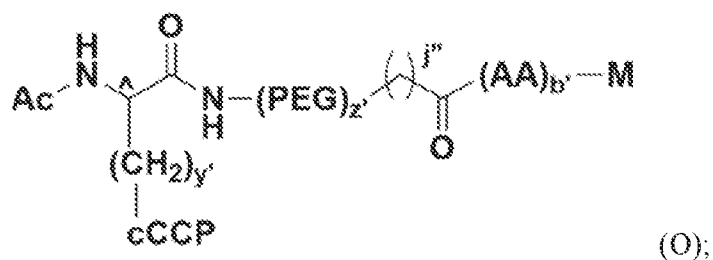
Formula (N):



wherein Ac, cCPP, Δ , y' , z' , j'' , $K^\#$, $X^{o'}$, $(AA)b'$, b' , and M are as defined herein.

[0237] In embodiments, the EEV comprises Formula (O):

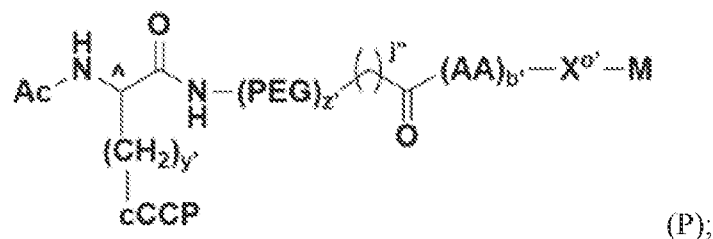
Formula (O):



5 wherein Ac, cCPP, Δ , y' , z' , j'' , $(AA)b'$, b' , and M are as defined herein.

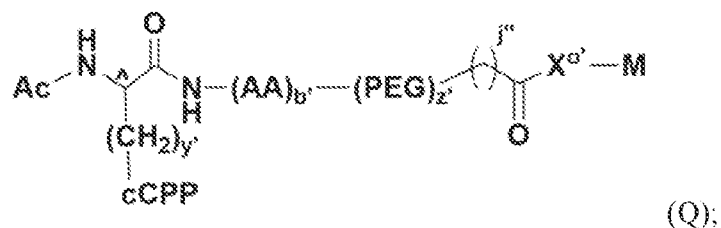
[0238] In embodiments, the EEV comprises Formula (P):

Formula (P):



wherein Ac, cCPP, Δ , y' , z' , j'' , $X^{o'}$, $(AA)b'$, b' , and M are as defined herein.

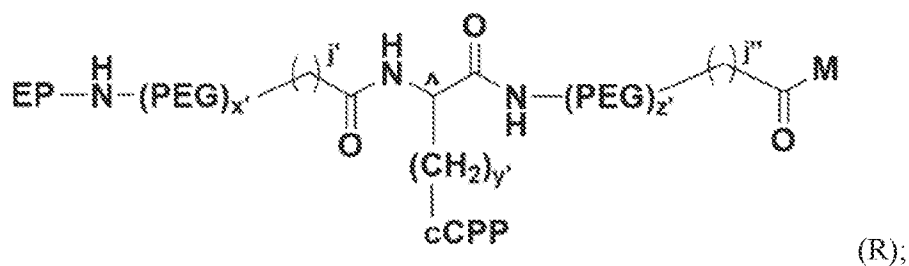
10 Formula (Q):



wherein Ac, cCPP, Δ , y' , z' , j'' , $X^{o'}$, $(AA)b'$, b' , and M are as defined herein.

[0239] In embodiments, the EEV comprises Formula (R):

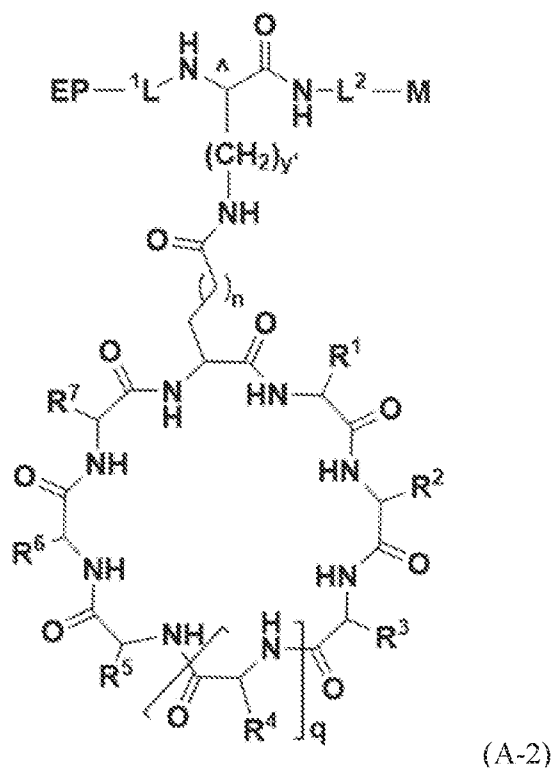
Formula (R):



wherein EP, cCPP, x' , j' , $\wedge$, v' , z' , j'' , and M are as defined herein.

[0240] In embodiments, the EEV can comprise Formula (A-2):

5 *Formula (A-2)*



wherein EP, L^1 , L^2 , M, y' , $\wedge$, n, q, R_1 , R_2 , R_3 , R_4 , R_5 , R_6 , and R_7 are as defined herein.

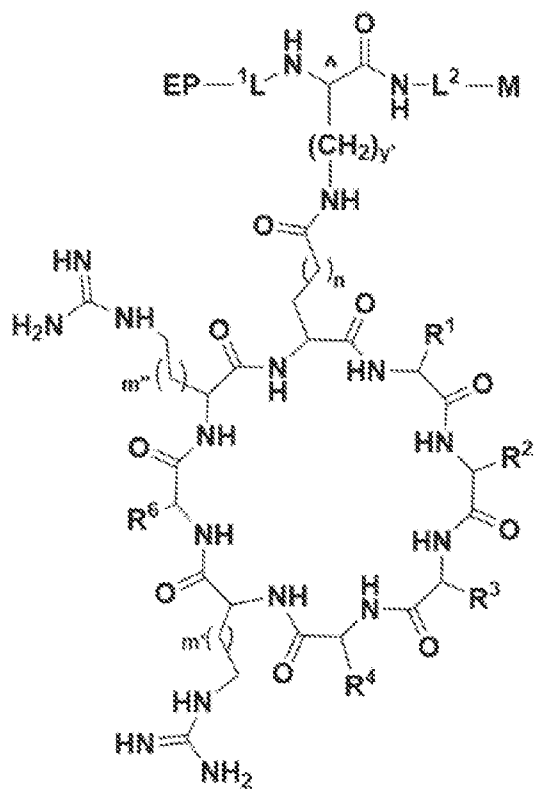
[0241] In embodiments, the EEV of Formula (A-2) can comprise a linker of Formula (B'), (BB'), (C'), (D'), (E') or (F').

10 [0242] In embodiments, the EEV of Formula (A-2) can comprise a linker of Formula (G'), (H'), (J'), (JJ'), (K'), (L'), or (M').

[0243] In embodiments, the EEV of Formula (A-2) can comprise a linker of Formula (R') or (B').

[0244] In embodiments, the EEV can comprise Formula (A-2a):

Formula (A-2a)



(A-2a)

wherein EP, L¹, L², M, y', ^, n, m', m'', R₁, R₂, R₃, R₄, and R₆, are as defined herein.

[0245] In embodiments, the EEV of Formula (A-2a) has a linker structure of Formula (B'), (BB'),

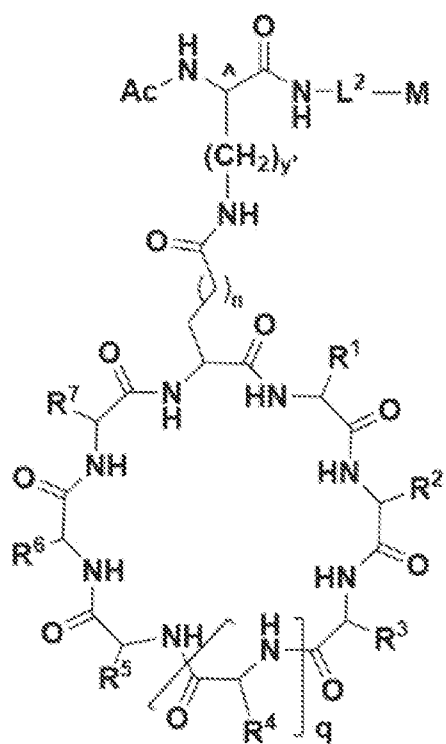
5 (C'), (D'), (E'), or (F').

[0246] In embodiments, the EEV of Formula (A-2a) can comprise a linker of Formula (G'), (H'), (J'), (JJ'), (K'), (L'), or (M').

[0247] In embodiments, the EEV of Formula (A-2a) can comprise a linker of Formula (R') or (B').

[0248] In embodiments, the EEV can comprise Formula (AA-2):

Formula (AA-2)



(AA-2)

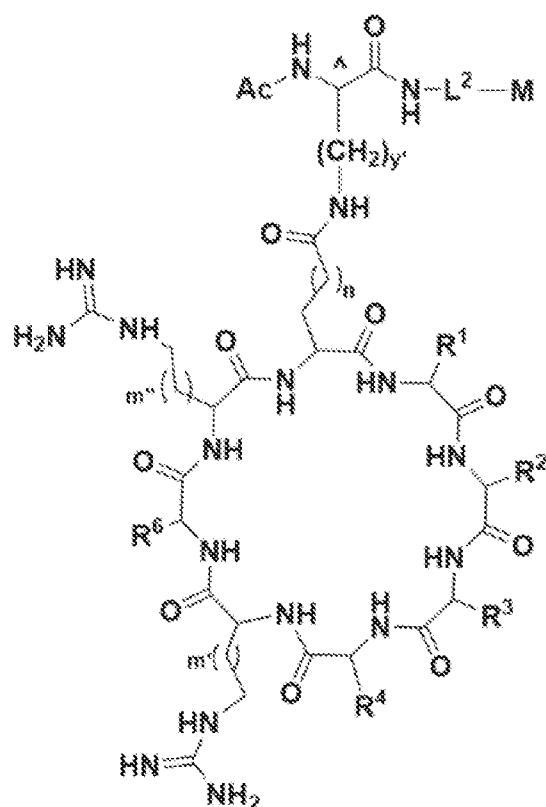
wherein Ac, L², M, y', ^, n, q, R₁, R₂, R₃, R₄, R₅, R₆, and R₇ are as defined herein.

[0249] In embodiments, the EEV of Formula (AA-2) can comprise a linker of Formula (N'), (O'),

5 (P'), or (Q').

[0250] In embodiments, the EEV can comprise Formula (AA-2a):

Formula (AA-2a)



(AA-2a)

wherein Ac, L², M, y', ^Δ, n, m', m'', R₁, R₂, R₃, R₄, and R₆, are as defined herein.

[0251] In embodiments, the EEV of Formula (AA-2a) can comprise a linker of Formula (N'), (O'), (P'), or (Q').

[0252] In embodiments, the EEV can be selected from:

Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-K(Ac-Bip)-OH
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-OH
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-OH
Ac-KKK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RK-Nal-OH
Ac-KK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-KRK-Nal-OH
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-KKKRK-PEG ₁₂ -Nal-OH
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-OH

Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-OH

Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -rk-nal-OH

Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkrk(Ac-nal)-OH
--

Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkr-nal-OH

[0253] In embodiments, the EEV can be selected from:

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -Nal-OH
--

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRGQ])-PEG ₁₂ -Nal-OH
--

[0254] In embodiments, the EEV is:

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Nal-PEG ₂ -Nal-OH
--

5

[0255] In embodiments, the EEV can be selected from:

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -dNal-OH

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -Nal-OH
--

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fffrrrrQ])-PEG ₁₂ -dNal-OH

[0256] In embodiments, the EEV is:

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -dNal-OH

10

[0257] In embodiments, the EEV is:

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -dNal-OH

[0258] In embodiments, the EEV can be selected from:

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH
--

Ac-KKKRK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH
--

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-OH
--

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH

[0259] In embodiments, the EEV can be selected from:

cyclo[FGFGRGRQ]-Lys(Bip-KRKKKP-Ac)-OH

cyclo[FGFGRGRQ]-Lys(PKKKRK-Bip-Ac)-OH

[0260] In embodiments, the EEV can be selected from:

Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-OH

Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-OH

Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-Nal-OH
--

Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-Nal-OH
--

[0261] In embodiments, the EEV can be selected from:

Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG12-Nal-OH

5

[0262] In embodiments, the EEV can be selected from:

Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG12-Nal-OH
--

[0263] In embodiments, the EEV can be selected from:

Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG12-Nal-OH
--

10 [0264] In embodiments, the EEV can be selected from:

Ac-RBRRBR-PEG2-K(cyclo[FfFGRGRQ])-PEG12-Nal-OH
--

Ac-RBRRBR-PEG2-K(cyclo[FfFRRRRQ])-PEG12-Nal-OH
--

Ac-RBRRBR-PEG2-K(cyclo[FfFRrRrQ])-PEG12-Nal-OH and
--

[0265] In embodiments, the EEV can be selected from:

Ac-rbrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-OH
--

[0266] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₁₂ -Nal-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRGQ])-PEG ₁₂ -Nal-OH

15

[0267] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-OH

[0268] In embodiments, the EEV is selected from:

Ac-PKKKRKV-K(cyclo[fFRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FAFARARQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG ₁₂ -Nal-OH and

[0269] In embodiments, the EEV is selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG ₁₂ -Nal-OH
--

5 [0270] In embodiments, the EEV is selected from:

Ac-RBRRBR-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRRRRQ])-PEG ₁₂ -Nal-OH
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRrRrQ])-PEG ₁₂ -Nal-OH and
Ac-rbrrbr-PEG ₂ -K(cyclo[fGfGrGrQ])-PEG ₁₂ -Nal-OH

[0271] In embodiments, the EEV is selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG ₁₂ -dNal-OH
Ac-pkkkrkv-PEG ₂ -k(cyclo[fffrrrQ])-PEG ₁₂ -dNal-OH
Ac-pkkkrkv-PEG ₂ -k(cyclo[FGFRRRRQ])-PEG ₁₂ -dNal-OH and
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -dNal-OH

[0272] In embodiments, the EEV is:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-Bip-OH.

[0273] In embodiments, the EEV is:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-C₆-OH.

[0274] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -F-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -W-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Y-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -V-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -I-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -L-OH and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -H-OH

[0275] In embodiments, the EEV is:

5 Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-G-OH.

[0276] In embodiments, the EEV is:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-Nal-PEG₂-Nal-OH.

[0277] In embodiments, the EEV is selected from:

Ac-KKK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-OH
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-OH
Ac-KKK-PEG ₂ -K(cyclo[Ff-Nal-GrGrQ])-PEG ₁₂ -RK-Nal-OH
Ac-KK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKK-Nal-OH
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-OH and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-OH

[0278] In embodiments, the EEV is selected from:

Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-OH
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-OH and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-OH

10 [0279] In embodiments, the EEV is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-OH and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH

[0280] In embodiments, the EEV is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH and
--

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-OH
--

[0281] In embodiments, the EEV is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-OH and
--

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-OH
--

[0282] In embodiments, the EEV is:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-OH

[0283] In embodiments, the EEV is selected from:

Ac-KKK-PEG ₂ -K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-OH
--

Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-OH and
--

Ac-K(cyclo[FGFGRGRQ])-KKKRK-PEG ₁₂ -Nal-OH

[0284] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Nal)-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Bip)-OH and

Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkrk(Ac-nal)-OH

5 [0285] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-W)-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Y)-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-I)-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-L)-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-H)-OH and

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-F)-OH

[0286] In embodiments, the EEV is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH
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Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BHRHR-OH and

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRHRH-OH

[0287] In embodiments, the EEV is:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RFQILY-PEG ₂ -OH.
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[0288] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -R-PEG ₄ -OH
--

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -F-PEG ₄ -OH
--

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RRF-PEG ₄ -OH
--

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RFR-PEG ₄ -OH
--

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RGRRG-PEG ₄ -OH
--

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RFRRF-PEG ₄ -OH
--

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RBRRBR-PEG ₄ -OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -KKKRK-PEG ₄ -OH and
--

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

[0289] In embodiments, the EEV is:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₂-RF-PEG₄-K(N₃)-NH₂.

[0290] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₄ -RF-PEG ₄ -OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -RF-PEG ₄ -OH and

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -RF-PEG ₄ -OH

5 [0291] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH and

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH

[0292] In embodiments, the EEV is:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₂ -Bip-OH.
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[0293] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-OH

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-OH and

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-OH

[0294] In embodiments, the EEV is:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₈-R-Bip-OH.

10 [0295] In embodiments, the EEV is:

Ac-PKKKRKG-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-RB-Bip-RB-OH.

[0296] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(KKKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂
Ac-KKKRK-PEG ₂ -K(RRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂
Ac-RRRBR-PEG ₂ -K(KKKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂ and
Ac-KRK-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂

[0297] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-RF-PEG ₄ -OH and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-RF-PEG ₄ -OH

[0298] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-GRF-PEG ₄ -OH and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-BRF-PEG ₄ -OH

5 [0299] In embodiments, the EEV is:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-GGE-OH or
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-GGE-NH ₂

[0300] In embodiments, the EEV is:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-GGRRGRRG-OH.

[0301] In embodiments, the EEV is:

Ac-PKKKRKV-PEG₂-K(cyclo[FfFGRGRQ])-PEG₂-RFQILYBRBRB-OH.

10 [0302] In embodiments, the EEV is:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₈-RF-OH.

[0303] In embodiments, the EEV is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-OH and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-OH

[0304] In embodiments, the EEV is:

Ac-PKKKRKG-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-RB-Bip-RB-OH.

15 [0305] In embodiments, the EEV is selected from:

Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKKR-K(Ac-Nal)-OH;
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKR-K(Ac-Nal)-OH; and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KR-K(Ac-Nal)-OH.

[0306] In embodiments, the EEV is selected from:

5 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKKR-K(Ac-dNal)-OH;
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKR-K(Ac-dNal)-OH; and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KR-K(Ac-dNal)-OH.

[0307] In embodiments, the EEV is selected from:

10 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeK-N^αMeK-N^αMeR-N^αMeK(Ac-Nal)-OH;
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeK-N^αMeR-N^αMeK(Ac-Nal)-OH; and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeR-N^αMeK(Ac-Nal)-OH.

[0308] In embodiments, the EEV is selected from:

15 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeK-N^αMeK-N^αMeR-N^αMeK(Ac-dNal)-OH;
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeK-N^αMeR-N^αMeK(Ac-dNal)-OH; and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeR-N^αMeK(Ac-dNal)-OH.

[0309] In embodiments, the EEV is selected from:

Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKKR-k(N₃)-NH₂
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKR-k(N₃)-NH₂ and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KR- k(N₃)-NH₂.

20 [0310] In embodiments, the EEV is selected from:

Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKKR-OH
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKR-OH and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KR-OH.

[0311] In embodiments, the EEV is selected from:

25 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂- N^αMe K-N^αMeK-N^αMeK-N^αMeR- k(N₃)-NH₂
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂- N^αMe K-N^αMeK-N^αMeR-k(N₃)-NH₂ and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeR- k(N₃)-NH₂.

[0312] In embodiments, the EEV is selected from:

30 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂- N^αMe K-N^αMeK-N^αMeK-N^αMeR-OH
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂- N^αMe K-N^αMeK-N^αMeR-OH and
 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-N^αMeK-N^αMeR-OH.

[0313] In embodiments, the EEV is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KR-Nal-OH and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-OH

[0314] In embodiments, the EEV is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N α MeK-N α MeK-N α MeK-N α MeR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N α MeK-N α MeK-N α MeR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N α MeK-N α MeK-N α MeR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N α MeK-N α MeR-Nal-OH and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -NMeK-NMeK-NMeR-Nal-OH

[0315] In embodiments, the EEV is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BHRHR-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRHRH-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-OH and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-OH

[0316] In embodiments, the EEV is selected from:

cyclo[FGFGRGRQ]-Lys(Bip-KRKKKP-Ac)-OH and
cyclo[FGFGRGRQ]-Lys(PKKKRK-Bip-Ac)-OH

5 [0317] In embodiments, the EEV is selected from:

Ac-pkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -OH and
--

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -K(N ₃)-NH ₂

[0318] In embodiments, the EEV is selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrrQ])-PEG ₁₂ -dNal-OH
--

Ac-pkkkrkv-PEG ₂ -k(cyclo[fffrrrrQ])-PEG ₁₂ -dNal-OH and
--

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -dNal-OH
--

[0319] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-KKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
--

Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
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Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
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Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH

Ac-KKKRK-PEG ₄ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH

Ac-KKKRK-PEG ₈ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH

Ac-KKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
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Ac-KRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH

Ac-RK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
--

Ac-RF-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
--

5 [0320] In embodiments, the EEV is selected from:

Ac-rbrrrbr-PEG₂-K(cyclo[fGfGrGrQ])-PEG₁₂-Nal-OH.

[0321] In embodiments, the EEV is selected from:

Ac-pkkkrkv-PEG₂-k(cyclo[fGfrrrrQ])-PEG₁₂-Nal-OH.

[0322] In embodiments, the EEV is selected from:

10 cyclo[FGFGRGRQ]-K(Bip-KRKKKP-Ac)-OH; and
cyclo[FGFGRGRQ]-K(PKKKRK-Bip-Ac)-OH.

[0323] In embodiments, the EEV is selected from:

Ac-KRK-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH

Ac-KRK-PEG ₂ -K(KRKIL-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH

Ac-KRKIL-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
Ac-KRK-PEG ₂ -K(KRKF-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
Ac-KRKF-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH

[0324] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
Ac-KKKRK-PEG ₂ -K(RBRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
Ac-RBRRBR-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH

[0325] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(RFRRF-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
Ac-RFRRF-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH

5

[0326] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(RIRRI-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
Ac-RIRRI-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH

[0327] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -FGF-K(cyclo[RGRGRGRQ])-PEG ₁₂ -OH

10 [0328] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[RGRGRGRQ])-PEG ₁₂ -dNaI-OH
--

[0329] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -R-OH

[0330] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH

15

[0331] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₂ -RF-PEG ₄ -F-OH

[0332] In embodiments, the EEV is selected from:

Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH
--

[0333] In embodiments, the EEV is selected from:

Ac-KKFRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH

Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH

Ac-KFK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH

5 [0334] In embodiments, the EEV is selected from:

Ac-RF-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
--

Ac-KKFRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

[0335] In embodiments, the EEV is selected from:

Ac-KKFRK-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
--

10 [0336] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGRGFGRQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-KKKRK-PEG ₂ -K(cyclo[FRFRFRFQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-KKKRK-PEG ₂ -K(cyclo[FGFLRLRQ])-PEG ₂ -RF-PEG ₄ -OH

Ac-KKKRK-PEG ₂ -K(cyclo[FGFVRVRQ])-PEG ₂ -RF-PEG ₄ -OH

[0337] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -KRR-PEG ₄ -OH
--

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -KKRR-PEG ₄ -OH

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -KKKRR-PEG ₄ -OH
--

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -KRK-PEG ₄ -OH
--

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -KKRK-PEG ₄ -OH

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -KKKRK-PEG ₄ -OH
--

[0338] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH

[0339] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH
Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH
Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH

[0340] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
Ac-KKKRK-PEG ₄ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
Ac-KKKRK-PEG ₈ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH

5

[0341] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
Ac-KRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
Ac-RK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH
Ac-RF-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH

[0342] In the EEV sequences provided, lysine can include a protecting group. In embodiments, the protecting group is a trifluoroacetyl (Tfa) group. In embodiments, the protecting group is an acetyl (Ac) group. It is understood that other protecting group can also be used and that the protecting group can be removed after the EEV is conjugated to a cargo. In embodiments, the EEVs are deprotected after conjugation to the PMOs.

10

[0343] Examples of EEV structures before conjugation to a cargo are shown in **FIGs. 6-12**.

Exocyclic Peptides

[0344] The exocyclic peptide (EP) can comprise from 2 to 10 amino acid residues e.g., 2, 3, 4, 5, 6, 7, 8, 9, or 10 amino acid residues. The EP can comprise 2 to 8 amino acid residues or 2 to 6

15

amino acid residues. In embodiments, the EP comprises 2 amino acid residues. In embodiments, the EP comprises 3 amino acid residues. In embodiments, the EP comprises 4 amino acid residues. In embodiments, the EP comprises 5 amino acid residues. In embodiments, the EP comprises 6 amino acid residues. In embodiments, the EP comprises 7 amino acid residues. In embodiments, the EP comprises 8 amino acid residues.

[0345] The amino acids in the EP can have D or L stereochemistry. The amino acid residues of the EP may all be D-amino acids. The amino acid residues of the EP may all be L-amino acids. The amino acid residues of the EP may be a combination of D-amino acids and L-amino acids.

[0346] The exocyclic peptide can be acylated at the N-terminus (Ac-EP). It is understood that EP and Ac-EP can be used interchangeably throughout the application. For example, the EP can comprise Ac-PKKKRKV.

[0347] Each amino acid in the exocyclic peptide may be a natural or non-natural amino acid. The term “non-natural amino acid” refers to an organic compound that is a congener of a natural amino acid in that it has a structure similar to a natural amino acid so that it mimics the structure and reactivity of a natural amino acid. The non-natural amino acid can be a modified amino acid, and/or amino acid analog, that is not one of the 20 common naturally occurring amino acids or the rare natural amino acids selenocysteine or pyrrolysine. Non-natural amino acids can also be the D-isomer of the natural amino acids. Non-natural amino acids can also be the D-isomer of the natural amino acids. Amino acids and/or amino acid residues may be referred to using their full name, their conventional three letter abbreviation, or their conventional one letter abbreviation. When using the one letter abbreviation, capital letters indicate L amino acids or residues, and lower case letters indicate D amino acids or residues. For example, arginine may be referred to as Arg, R, or r.

[0348] The EP can comprise at least one amino acid residue that is positively charged at physiologically relevant pH values. In embodiments, the EP comprises at least one amino acid residue comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof. In embodiments, the EP comprises at least one amino acid residue comprising a guanidine group, or a protonated form thereof. In embodiments, the EP comprises at least one amino acid residue comprising a terminal amine or a protonated form thereof. In embodiments, the EP comprises at least one amino acid residue comprising an imidazole, or a protonated form thereof. Protonated forms can mean salt thereof throughout the disclosure.

[0349] The EP can comprise from 1 to 5 amino acids residues comprising a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof. The EP can comprise from 1 to 4 amino acids residues comprising a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof. The EP can
5 comprise from 1 to 3 amino acids residue comprising a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof. The EP can comprise 1 or 2 amino acids residue comprising a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof. The EP can comprise 2 or 3 amino acids residue comprising a side chain comprising a guanidine group, a terminal amine, an imidazole, or a
10 protonated form thereof. The EP can comprise from 2 to 4 amino acids residue comprising a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof.

[0350] In embodiments, the amino acid comprising a guanidine group is arginine. In embodiments, the amino acid comprising a terminal amine is lysine. In embodiments, the amino acid comprising
15 an imidazole is histidine.

[0351] The EP can comprise 1, 2, 3, 4, or 5 amino acids residues comprising a side chain comprising a guanidine group, or a protonated form thereof. The EP can comprise 1 amino acid residue comprising a side chain comprising a guanidine group, or a protonated form thereof. The EP can comprise amino acid residues comprising a side chain comprising a guanidine group, or a
20 protonated form thereof. The EP can comprise 3 amino acid residues comprising a side chain comprising a guanidine group, or a protonated form thereof. The EP can comprise 4 amino acid residues comprising a side chain comprising a guanidine group, or a protonated form thereof. The EP can comprise 5 amino acid residues comprising a side chain comprising a guanidine group, or a protonated form thereof. The amino acid residue comprising a side chain comprising
25 a guanidine group can be an arginine residue. The EP can comprise 1, 2, 3, 4, or 5 arginine residues. The EP can comprise 1 arginine residue. The EP can comprise 2 arginine residues. The EP can comprise 3 arginine residues. The EP can comprise 4 arginine residues. The EP can comprise 5 arginine residues.

[0352] The EP can comprise 1, 2, 3, 4, or 5 amino acid residues comprising a terminal amine, or
30 a protonated form thereof. The EP can comprise 1 amino acid residue comprising a terminal amine, or a protonated form thereof. The EP can comprise 2 amino acid residues comprising a terminal

amine, or a protonated form thereof. The EP can comprise 3 amino acid residues comprising a terminal amine, or a protonated form thereof. The EP can comprise 4 amino acid residues comprising a terminal amine, or a protonated form thereof. The EP can comprise 5 amino acid residues comprising a terminal amine, or a protonated form thereof. The amino acid with a terminal amine can be lysine. The EP can comprise 1, 2, 3, 4, or 5 lysine residues. The EP can comprise 1 lysine residue. The EP can comprise 2 lysine residues. The EP can comprise 3 lysine residues. The EP can comprise 4 lysine residues. The EP can comprise 5 lysine residues. The amino group on the side chain of each lysine residue can be substituted with a protecting group, including, for example, trifluoroacetyl (-COCF₃), allyloxycarbonyl (Alloc), 1-(4,4-dimethyl-2,6-dioxocyclohexylidene)ethyl (Dde), or (4,4-dimethyl-2,6-dioxocyclohex-1-ylidene-3)-methylbutyl (ivDde) group. The amino group on the side chain of each lysine residue can be substituted with a trifluoroacetyl (-COCF₃) group. The protecting group can be included to enable amide conjugation. The protecting group can be removed after the EP is conjugated to a cCPP.

[0353] The EP can comprise 1, 2, 3, 4, or 5 amino acid residues comprising an imidazole, or a protonated form thereof. The EP can comprise 1 amino acid residue comprising an imidazole, or a protonated form thereof. The EP can comprise 2 amino acid residues comprising an imidazole, or a protonated form thereof. The EP can comprise 3 amino acid residues comprising an imidazole, or a protonated form thereof. The EP can comprise 4 amino acid residues comprising an imidazole, or a protonated form thereof. The EP can comprise 5 amino acid residues comprising an imidazole, or a protonated form thereof. The amino acid comprising an imidazole can be histidine. The EP can comprise 1, 2, 3, 4, or 5 histidine residues. The EP can comprise 1 histidine residue. The EP can comprise 2 histidine residues. The EP can comprise 3 histidine residues. The EP can comprise 4 histidine residues. The EP can comprise 5 histidine residues.

[0354] The EP can comprise 1, 2, 3, or 4 amino acid residues with an uncharged side chain. The uncharged side chain can comprise a hydrophobic side chain. The EP can comprise 1, 2, 3, or 4 amino acid residues with a hydrophobic side chain. The EP can comprise 1, 2, 3, or 4 amino acid residues with an uncharged hydrophobic side chain. The EP can comprise 1 amino acid residue with an uncharged hydrophobic side chain. The EP can comprise 2 amino acid residues with an uncharged hydrophobic side chain. The EP can comprise 3 amino acid residues with an uncharged hydrophobic side chain. The EP can comprise 4 amino acid residues with an uncharged hydrophobic side chain. The amino acid residue with an uncharged hydrophobic side chain can be

selected from valine, proline, β -alanine, and glycine. The amino acid residue with a hydrophobic side chain can be valine or proline. The amino acid residue with a hydrophobic side chain can be valine. The amino acid residue with a hydrophobic side chain can be proline. The amino acid residue with a hydrophobic side chain can be β -alanine. The amino acid residue with a hydrophobic side chain can be glycine.

[0355] The EP can comprise at least one positively charged amino acid residue and at least one uncharged hydrophobic residue. The EP can comprise two positively charged amino acid residues and one uncharged hydrophobic residue. The EP can comprise three positively charged amino acid residues and one uncharged hydrophobic residue. The EP can comprise four positively charged amino acid residues and one uncharged hydrophobic residue. The EP can comprise two positively charged amino acid residues and two uncharged hydrophobic residues. The EP can comprise three positively charged amino acid residues and two uncharged hydrophobic residues. The EP can comprise four positively charged amino acid residues and two uncharged hydrophobic residues.

[0356] The EP can comprise at least one lysine residue and at least one arginine residue. The EP can comprise 2, 3, 4, or 5 lysine residues and/or arginine residues. The EP can comprise 2 lysine and/or arginine residues. The EP can comprise 3 lysine and/or arginine residues. The EP can comprise 4 lysine and/or arginine residues. The EP can comprise 5 lysine and/or arginine residues.

[0357] The EP can comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof. The EP can comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), or combinations thereof. The EP can comprise one proline (P). The EP can comprise one valine (V). The EP can comprise one glycine (G). The EP can comprise one or more lysine (K). The EP can comprise one lysine (K). The EP can comprise two lysine (K). The EP can comprise three lysine (K). The EP can comprise four lysine (K). The EP can comprise one or more arginine (R). The EP can comprise one arginine (R). The EP can comprise two arginine (R). The EP can comprise three arginine (R). The EP can comprise four arginine (R). The EP can comprise five arginine (R). The EP can comprise six arginine (R). The EP can comprise one or more histidine (H). The EP can comprise one histidine (H). The EP can comprise two histidine (H). The EP can comprise three histidine (H). The EP can comprise four histidine (H). The EP can comprise five histidine (H). The EP can

comprise six histidine (H). The EP can comprise one or more β -alanine (B). The EP can comprise two β -alanine (B). The EP can comprise two β -alanine (B).

[0358] The EP can comprise KK, KR, RR, RK, RF, HH, HK, HR, RH, KKK, KFK, KGK, KBK, KBR, KRK, KRR, RKK, RRR, RHR, RRV, FRR, RFR, RBR, KKH, KHK, HKK, HRR, HRH, HHR, HBH, HHH, HHHH, PKKK, KHKK, KKHK, KKKH, KHKH, HKHK, KKKK, KKRK, KRKK, KRRK, KRKF, RKKR, RRRR, RBRB, BRBR, HRHR, RHRH, HBHB, BHBH, KGKK, KKGK, HBHBH, RBHBH, BRBRB, HBRBH, BHRHR, BRHRH, HBKBH, RRRRR, KKKKK, KKKRK, RKKKK, KRKKK, KKRKK, KKKKR, KKFRK, KBKKB, KRKIL, RBFBR, RBRBR, RBHBR, RFRRF, RIRRI, RKKKKG, KRKKKG, KKRKKG, KKKKRG, KKKRKG, RKKKKKB, KRKKKB, KKRKKB, KKKKRB, KKKRKV, RRRRRR, HHHHHH, RHRHRH, HRHRHR, KRKKKP, PKKKRK, KRKRKR, RKRKRK, RBRBRB, RBRRBR, KBKKB, PKKKRKV, PGKKRKV, PKGKRKV, PKKGRKV, PKKKGKV, PKKKRGV or PKKKRKG, wherein B is beta-alanine. The EP can comprise PKKKRKV. The EP can comprise RR, RRR, RHR, RBR, RBRBR, RBRRBR, RBGRBR, RBRGBR, RBRRBG, RBHBR, or HBRBH,

wherein B is beta-alanine. The amino acids in the EP can have D or L stereochemistry.

[0359] The EP can comprise kk, kr, rr, rk, rf, hh, hk, hr, rh, kkk, kfk, kgk, kbk, kbr, krk, krr, rkk, rrr, rhr, rrv, frr, rfr, rbr, kkh, khk, hkk, hrr, hrh, hhr, hbb, hhh, hhhh, pkkk, khkk, kkhk, kkkh, khkh, hkhk, kkkk, kkrk, krkk, krrk, krkf, rkk, rrrr, rbrb, brbr, hrhr, rhrh, hbhb, bbbh, kgkk, kkgk, hbhbh, rbhbh, brbrb, hbrbh, bhrhr, brhrh, hbkbh, rrrrr, kkkkk, kkkrk, rkkkk, krkkk, kkrkk, kkkkr, kkrfk, kbkbk, krkil, rbfbr, rbrbr, rbhbr, rfrrf, rirri, rkkkkk, krkkkk, kkrkkk, kkkkrg, rkkkkb, kkkkkb, kkkkkr, kkkkrv, rrrrrr, hhhhhh, rhrhrh, hrhrhr, krkkkp, pkkkrk, krkrkr, rkrkrk, rbrbrb, rbrrbr, kbkbkb, pkkkrkv, pgkkrkv, pkgkrkv, pkkgrkv, pkkkgkv, pkkkrgv or pkkkrkg.

[0360] The EP can consist of KK, KR, RR, RK, RF, HH, HK, HR, RH, KKK, KFK, KGK, KBK, KBR, KRK, KRR, RKK, RRR, RHR, RRV, FRR, RFR, RBR, KKH, KHK, HKK, HRR, HRH, HHR, HBH, HHH, HHHH, PKKK, KHKK, KKHK, KKKH, KHKH, HKHK, KKKK, KKRK, KRKK, KRRK, KRKF, RKKR, RRRR, RBRB, BRBR, HRHR, RHRH, HBHB, BHBH, KGKK, KKGK, HBHBH, RBHBH, BRBRB, HBRBH, BHRHR, BRHRH, HBKBH, RRRRR, KKKKK, KKKRK, RKKKK, KRKKK, KKRKK, KKKKR, KKFRK, KBKKB, KRKIL, RBFBR, RBRBR, RBHBR, RFRRF, RIRRI, RKKKKG, KRKKKG, KKRKKG, KKKKRG, KKKRKG, RKKKKKB, KRKKKB, KKRKKB, KKKKRB, KKKRKV, RRRRRR, HHHHHH, RHRHRH, HRHRHR, KRKKKP, PKKKRK, KRKRKR, RKRKRK, RBRBRB, RBRRBR,

KBKBKB, PKKKRKV, PGKKRKV, PKGKRKV, PPKGRKV, PKKKGKV, PKKKRGV or PKKKRKV, wherein B is beta-alanine. The EP can consist of PKKKRKV. The EP can consist of RR, RRR, RHR, RBR, RBRBR, RBRRBR, RBGRBR, RBRGBR, RBRRBG, RBHBR, or HBRBH, wherein B is beta-alanine. The amino acids in the EP can have D or L stereochemistry.

[0361] The EP can consist of kk, kr, rr, rk, rf, hh, hk, hr, rh, kkk, kfk, kgk, kbk, kbr, krk, krr, rkk, rrr, rhr, rrv, frf, rfr, rbr, kkh, khk, hkk, hrr, hrh, hhr, hbh, hhh, hhhh, pkkk, khkk, kkhk, kkkh, khkh, hkhk, kkkk, kkrk, krkk, krrk, krkf, rkk, rrrr, rrb, brbr, hrhr, rhrh, hbhb, bbbh, kgkk, kkgk, hbhb, rbhb, brbr, hbrb, bhrh, brhr, hbkb, rrrr, kkkk, kkrk, rkkk, krkk, kkrk, kkkk, kkrk, kbkb, krkl, rbf, rbr, rbbr, rrrf, rrr, rkkk, kkrk, kkkrg, kkkkb, kkrkb, kkkrb, kkrkv, rrrr, hhhhh, rhrh, hrhr, rkkp, pkkk, krkr, rkrk, rbrb, rrrr, kbkb, pkkk, pgkr, pgrk, pkkg, pkrv or pkrk.

[0362] The EP can comprise KK, KR, RR, RK, RF, KKK, KGK, KFK, KBK, KBR, KRK, KRR, RKK, RRR, KKKK, KKRK, KRKK, KRRK, KRKF, RKKR, RRRR, KGKK, KKGK, KKKKK, KKKRK, KBKBK, KKFRK, KRKIL, RFRRF, RIRRI, KKKRKV, PKKKRKV, PGKKRKV, PKGKRKV, PKKGRKV, PKKKGKV, PKKKRGV or PKKKRKG. The EP can comprise PKKKRKV, RR, RRR, RHR, RBR, RBRBR, RBHBR, or HBRBH, wherein B is beta-alanine.

[0363] The EP can comprise one or more $N^{uMe}K$ or $N^{uMe}R$ amino acid residues, or a combination thereof. The notation “ N^{uMe} ” indicates that the amino acid includes a methyl group on the N atom of the indicated amino acid. The EP can comprise $N^{uMe}K$ - $N^{uMe}R$; $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$; $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$; $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$; $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$; or $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$. The EP can consist of $N^{uMe}K$ - $N^{uMe}R$; $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$; $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$; $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$; $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$; or $N^{uMe}K$ - $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$ - $N^{uMe}R$ - $N^{uMe}K$.

[0364] The EP can comprise kk, kr, rr, rk, rf, kkk, kgk, kfk, kbk, kbr, krk, krr, rkk, rrr, kkkk, kkrk, krkk, krrk, krkf, rkkr, rrrr, kgkk, kkgk, kkkkk, kkkrk, kkbk, kkfrk, krkil, rfrf, rirri, kkkrv, pkkkrv, pgkkrv, pkgkrv, pkkgrv, pkkkgv, pkkkrgv, or pkkkrkg. The EP can comprise pkkkrv, rr, rrr, rhr, rbr, rbrbr, rbhbr, or hbrbh.

[0365] The EP can consist of KK, KR, RR, RK, RF, KKK, K GK, KFK, KBK, KBR, KRK, KRR, RKK, RRR, KKKK, KKRK, KRKK, KRRK, KRKF, RKKR, RRRR, KGKK, KKGK, KKKKK, KKKRK, KBKBK, KKFRK, KRKIL, RFRRF, RIRRI, KKKRKV, PKKKRKV, PGKKRKV, PKGKRKV, PKKGRKV, PKKKGKV, PKKKRGV or PKKKRKV. The EP can

consist of PKKKRKV, RR, RRR, RHR, RBR, RBRBR, RBHBR, or HBRBH, wherein B is beta-alanine.

[0366] The EP can consist of kk, kr, rr, rk, rf, kkk, kgk, kbk, kfk, kbr, krk, krr, rkk, rrr, kkkk, kkrk, krkk, krrk, krkf, rkk, rrrr, kgkk, kkgk, kkkkk, kkkrk, kbkbk, kkfrk, krkil, rfrf, rirri, kkkkrkv, pkkkrkv, pgkkkrkv, pkgkrkv, pkkgrkv, pkkkgkv, pkkkrkv, or pkkkrkg.

[0367] The EP can comprise one of the following sequences: HBH, HBHBH, or HBRBH. The EP can comprise HHRBFBR. The EP can comprise one of the following sequences: KBK, KBKKBK or KBR. The EP can comprise one of the following sequences: KGK or KGKK. The EP can comprise one of the following sequences: KKGK, KKKK, KKKKR, KKKRK, KKKRKG, or KKKRK. The EP can comprise one of the following sequences: KR, KRK, or KRKKK. The EP can comprise PGKKRKV. The EP can comprise one of the following sequences: PKGKRKV, PKK, PKKGRKV, PKKK, PKKKGKV, PKKKRGV, PKKKRKG, PKKRGV, PKKRKG, or PKKRKV. The EP can comprise one of the following sequences: RBHBH, RBHBR, RBR, RBRBR, or RBRBRBR. The EP can comprise one of the following sequences: RFR, or RHR. The EP can comprise RKKK. The EP can comprise one of the following sequences: RR, RRR, or RRRRFFF. The EP can comprise one of the following sequences: RK or RF. The EP can comprise one of the following sequences: KFK or KRKF. The EP can comprise one of the following sequences: KKFRK or KRKIL. The EP can comprise one of the following sequences: RFRRF or RIRRI. The EP can comprise PKKKRKV. B is beta-alanine.

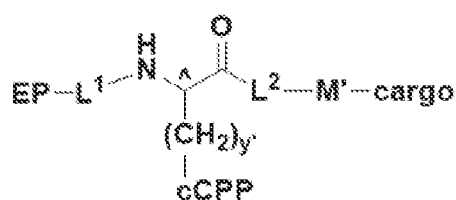
[0368] The EP can comprise an amino acid sequence identified in the art as a nuclear localization sequence (NLS). The EP can consist of an amino acid sequence identified in the art as a nuclear localization sequence (NLS). The EP can comprise an NLS comprising the amino acid sequence PKKKRKV. The EP can consist of an NLS comprising the amino acid sequence PKKKRKV.

[0369] The EP comprise from 2 to 10, 6 to 9, or 4 to 8 consecutive amino acid residues. The EP can comprise between 2 to 10, 6 to 9 or 4 to 8 amino acids, wherein not all amino acids are consecutive, for example, the amino acid residues of the EP may be separated by one or more components of the EEV, for example, the amino acid residues of an EP may be separated by a linker, the cCPP or a combination thereof, thereby forming a "split" EP.

EEV-cargo conjugate

[0370] In embodiments, the EEV can be conjugated to a cargo to form an EEV-cargo conjugate comprising the structure of Formula (A-1):

Formula (A-1):



5 , wherein

EP is a linear exocyclic peptide defined herein;

cCPP is a cyclic cell penetrating peptide defined herein;

L^1 and L^2 , are, independently, a linker arm;

^ indicates D- or L-stereochemistry;

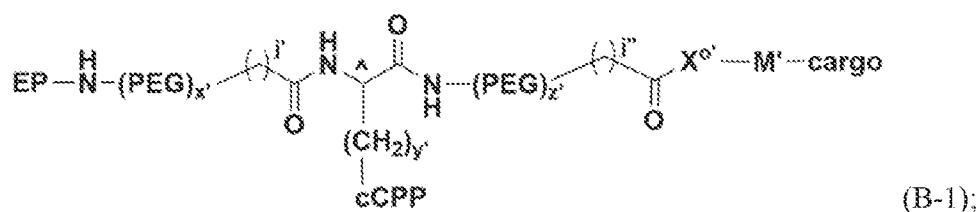
10 y' is an integer from 1 to 5;

M' is a bonding group defined herein; and

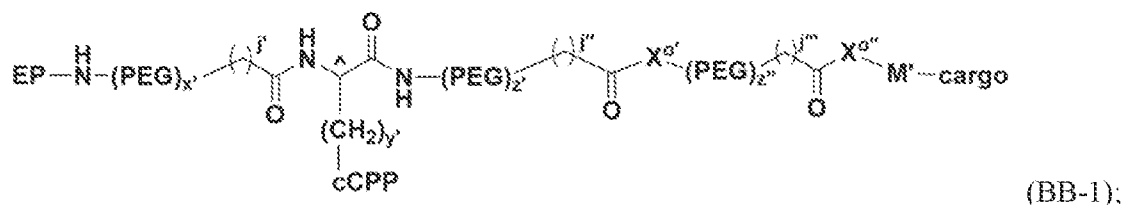
cargo is a peptide, an oligonucleotide, a small molecule, or any combinations thereof.

[0371] In embodiments, the compound Formula (A-1) can have a Formula selected from (B-1), (BB-1), (C-1), (D-1), (E-1) and (F-1):

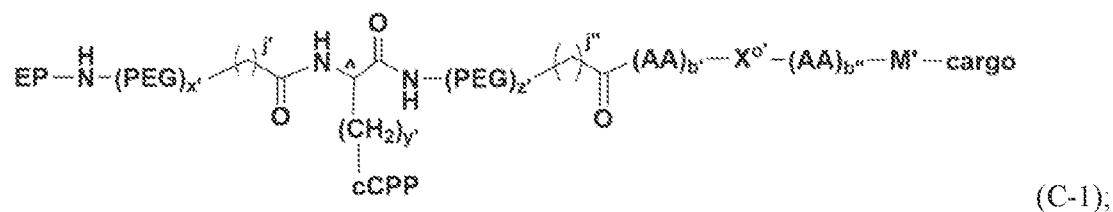
15 Formula (B-1):



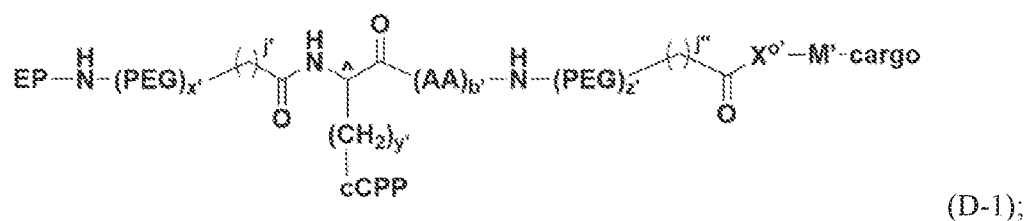
Formula (BB-1):



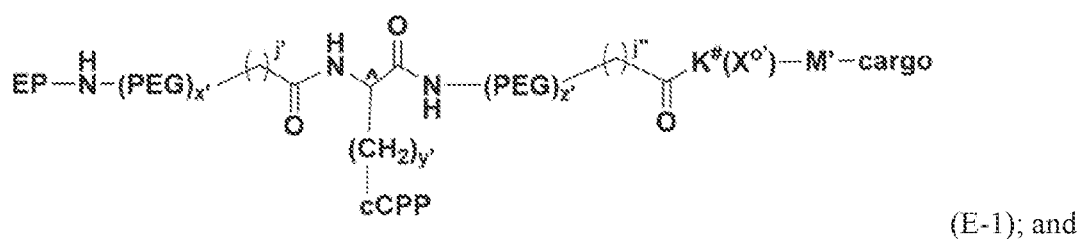
Formula (C-1):



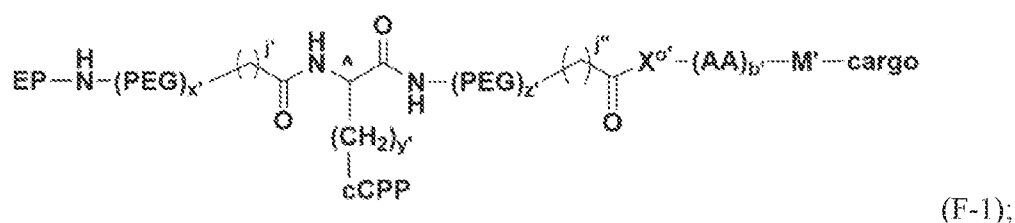
Formula (D-1):



5 Formula (E-1):



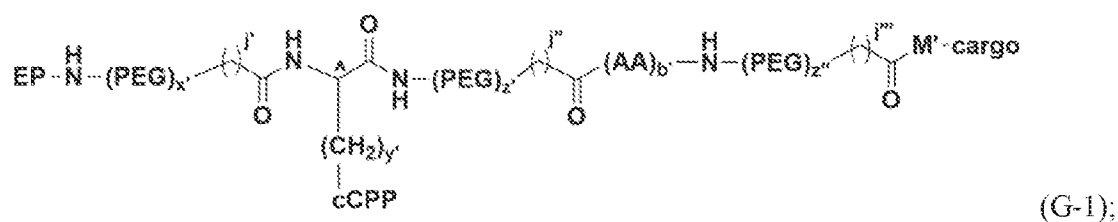
Formula (F-1):



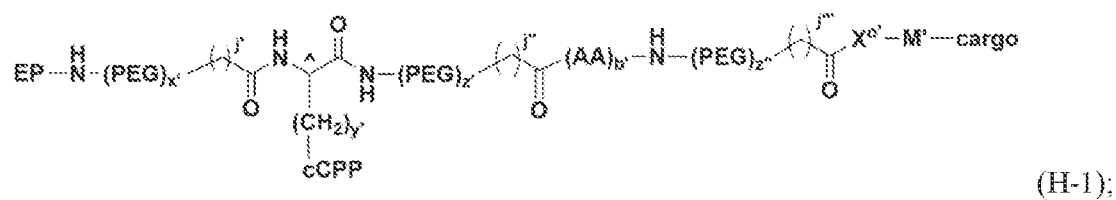
wherein EP, cCPP, x' , j' , $\wedge$, y' , z' , j'' , z'' , j''' , $X^{o'}$, $X^{o''}$, $K^{\#}$, $(\text{AA})_{b'}$, b' , M' , and cargo are as defined
10 herein.

[0372] In embodiments, the compound Formula (A-1) can have a Formula selected from (G-1), (H-1), (I-1), (J-1), (JJ-1), (K-1), (L-1), or (M-1):

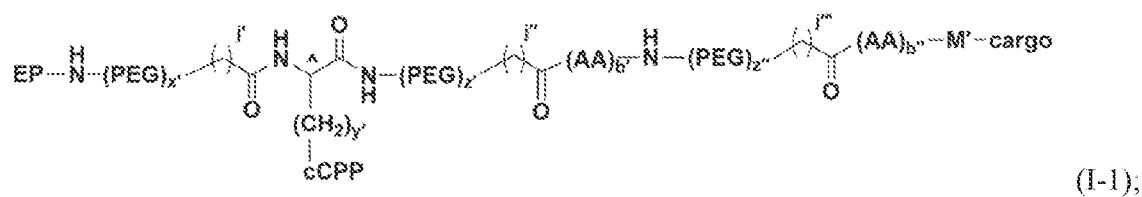
Formula (G-1):



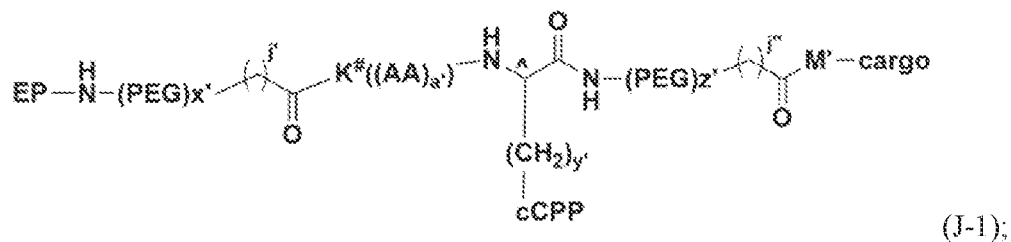
Formula (H-1):



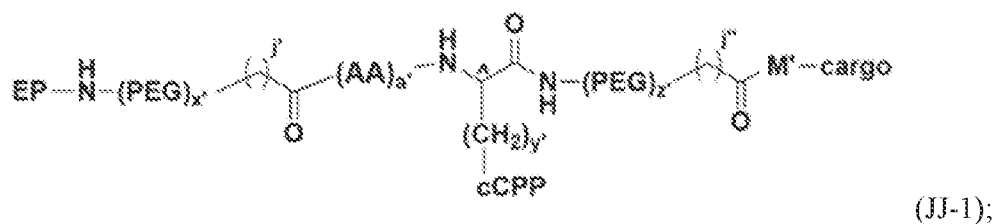
5 Formula (I-1):



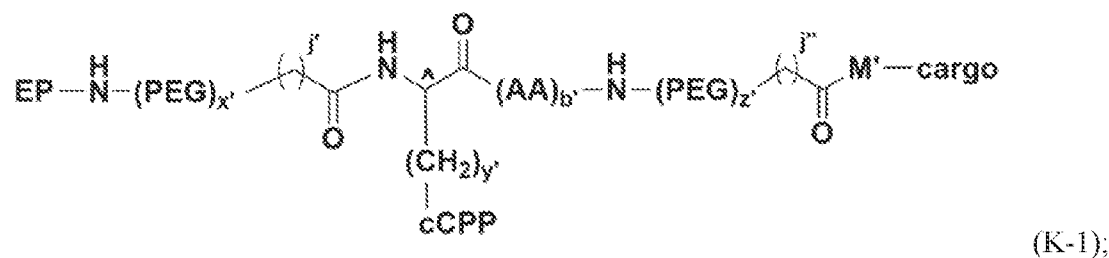
Formula (J-1):



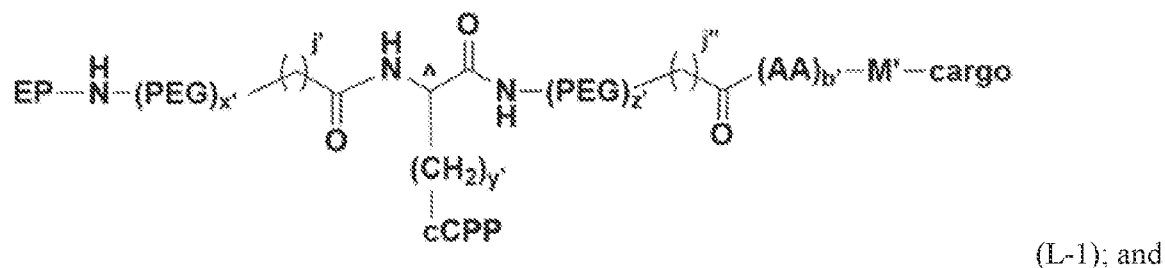
Formula (JJ-1):



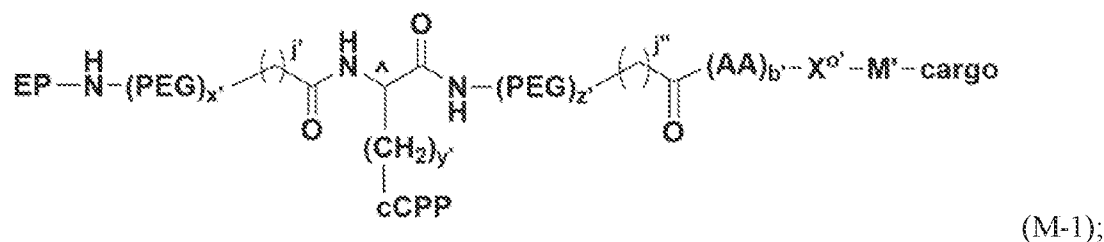
Formula (K-1):



Formula (L-1):



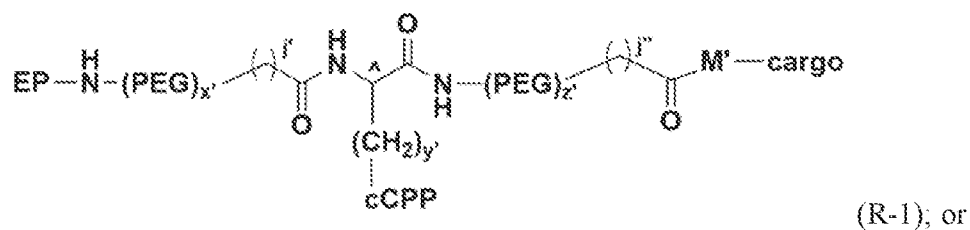
5 Formula (M-1):



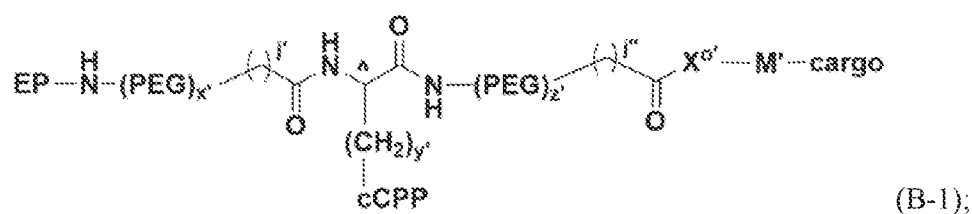
wherein EP, cCPP, x' , j' , y' , z' , j'' , z'' , j''' , $X^{o'}$, $K^\#$, $(\text{AA})_{a'}$, $(\text{AA})_{b'}$, $(\text{AA})_{b''}$, a' , b' , b'' , M' , and cargo are as defined herein.

[0373] In embodiments, the compound of Formula (A-1) can have a Formula (R-1) or (B-1):

10 Formula (R-1):



Formula (B-1):



wherein

x' is an integer from 0 to 12;

5 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

$\wedge$ indicates L- or D-stereochemistry;

y' is an integer from 1 to 5;

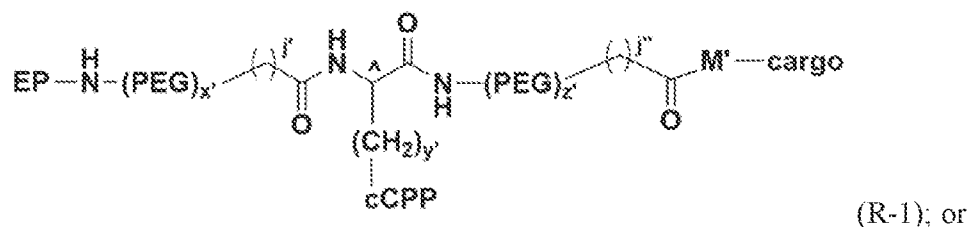
z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

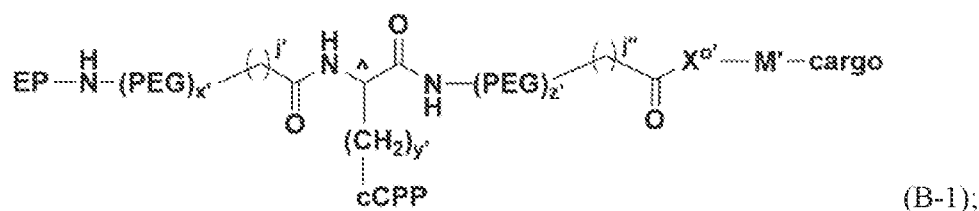
10 $X^{o'}$ comprises a hydrophobic component; and

[0374] cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof. In embodiments, the compound of Formula (A-1) can have a Formula (R-1) or (B-1):

Formula (R-1):



15 Formula (B-1):



wherein

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

20 $\wedge$ indicates D-stereochemistry;

y' is an integer from 1 to 5;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

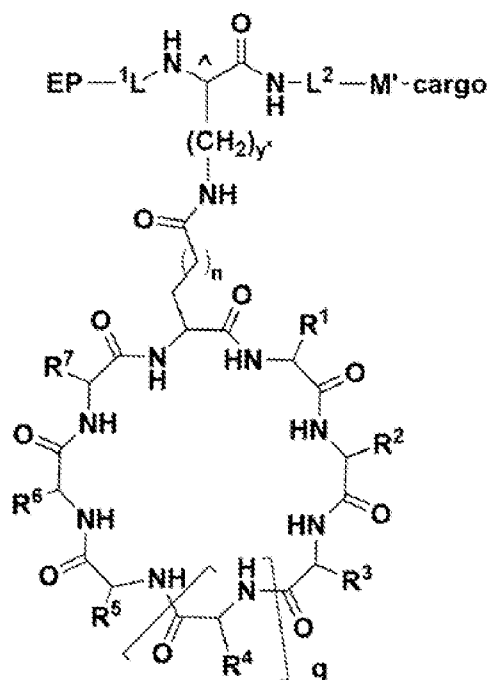
$X^{o'}$ comprises a hydrophobic component; and

cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof,

- 5 wherein EP comprises all D-amino acids; cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates cCPP to the linker.

[0375] In embodiments, the EEV-cargo conjugate has the structure of formula (A-3):

Formula (A-3):



(A-3)

wherein:

EP is a linear exocyclic peptide;

L^1 and L^2 are linker arms,

M' is a bonding group; and

y' is an integer from 1-5,

$\wedge$ indicates D- or L- stereochemistry;

R_1 , R_2 , and R_3 can each independently be H or an amino acid residue having a side chain comprising an aryl or heteroaryl group;

R_4 , R_5 , R_6 and R_7 are independently H or a side chain of an amino acid;

q is an integer from 1-4;

n is an integer from 1-4; and

cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

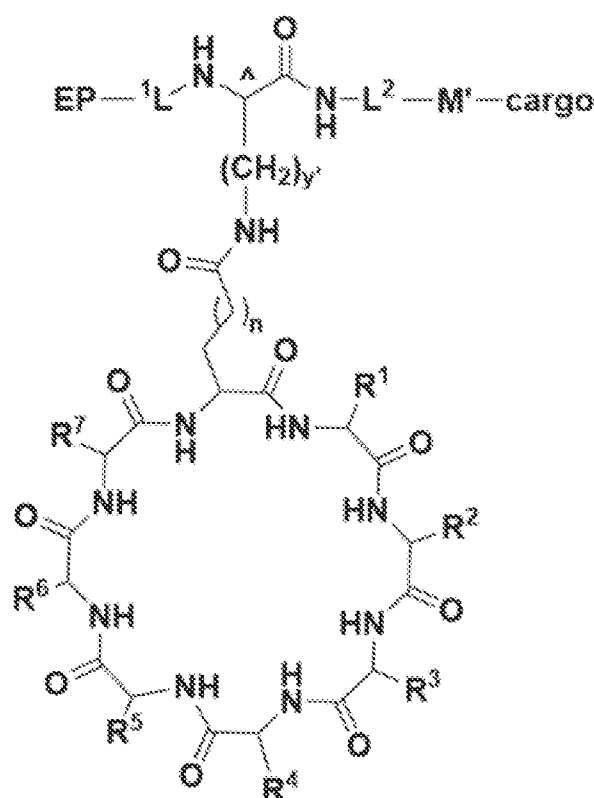
5 [0376] In embodiments, the EEV-cargo conjugate of Formula (A-3) has a linker structure of Formula (B'), (BB'), (C'), (D'), (E') or (F').

[0377] In embodiments, the EEV-cargo conjugate of Formula (A-3) has a linker structure of Formula (G'), (H'), (J'), (JJ'), (K'), (L'), or (M').

10 [0378] In embodiments, the EEV-cargo conjugate of Formula (A-3) has a linker structure of Formula (R') or (B').

[0379] In embodiments, the EEV-cargo conjugate has the structure of formula (A-3a):

Formula (A-3a)



(A-3a)

15 wherein EP, M' , L^1 , L^2 , $\wedge$, y' , n , R_1 , R_2 , R_3 , R_4 , R_5 , R_6 , and R_7 are as defined herein and cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

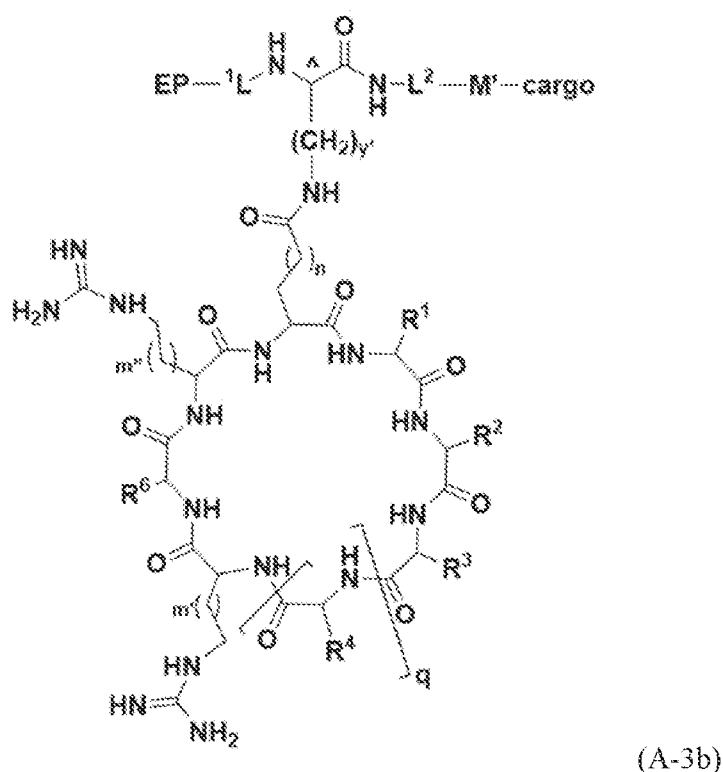
[0380] In embodiments, the EEV-cargo conjugate of Formula (A-3a) has a linker structure of Formula (B'), (BB'), (C'), (D'), (E'), or (F').

[0381] In embodiments, the EEV-cargo conjugate of Formula (A-3a) has a linker structure of Formula (G'), (H'), (J'), (JJ'), (K'), (L'), or (M').

5 [0382] In embodiments, the EEV-cargo conjugate of Formula (A-3a) has a linker structure of Formula (R') or (B').

[0383] In embodiments, the EEV-cargo conjugate has the structure of formula (A-3b):

Formula (A-3b):



10 wherein EP, L¹, L², M', ^, y', n, m', m'', q, R₁, R₂, R₃, R₄, and R₆ are as defined herein and cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

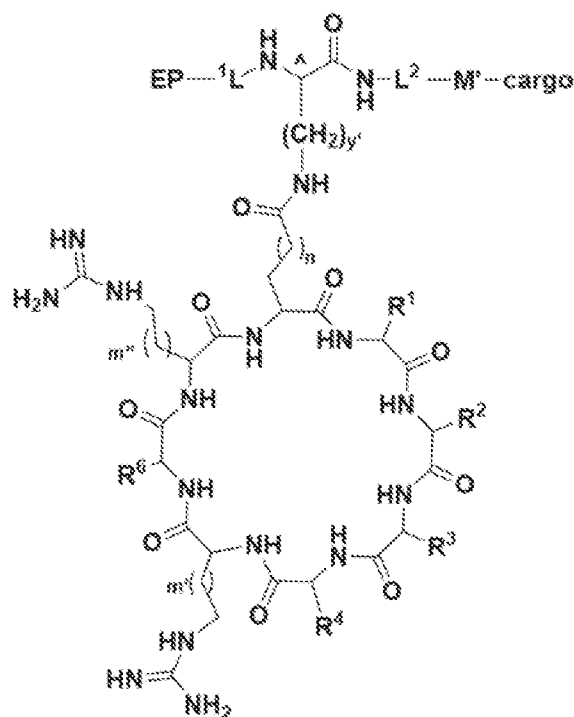
[0384] In embodiments, the EEV-cargo conjugate of Formula (A-3b) has a linker structure of Formula (B'), (BB'), (C'), (D'), (E'), or (F').

15 [0385] In embodiments, the EEV-cargo conjugate of Formula (A-3b) has a linker structure of Formula (G'), (H'), (J'), (JJ'), (K'), (L'), or (M').

[0386] In embodiments, the EEV-cargo conjugate of Formula (A-3b) has a linker structure of Formula (R') or (B').

[0387] In embodiments, the EEV-cargo conjugate has the structure of formula (A-3c):

Formula (A-3c):



wherein EP, L¹, L², M', ^, y', n, m', m'', R₁, R₂, R₃, R₄, and R₆ are as defined herein and cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

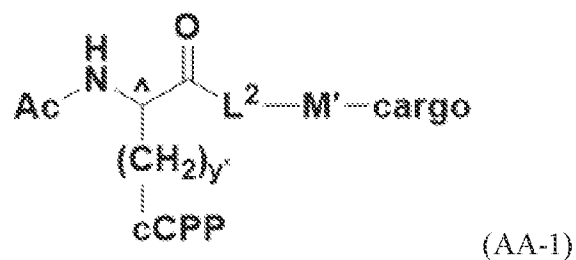
[0388] In embodiments, the EEV-cargo conjugate of Formula (A-3c) has a linker structure of Formula (B'), (BB'), (C'), (D'), (E'), or (F').

[0389] In embodiments, the EEV-cargo conjugate of Formula (A-3c) has a linker structure of Formula (G'), (H'), (J'), (JJ'), (K'), (L'), or (M').

[0390] In embodiments, the EEV-cargo conjugate of Formula (A-3c) has a linker structure of Formula (R') or (B').

[0391] In embodiments, the EEV can be conjugated to a cargo to form an EEV-cargo conjugate comprising the structure of Formula (AA-1):

Formula (AA-1):



wherein:

cCPP is a cyclic cell penetrating peptide;

Ac is acetyl;

L² is a linker arm;

^ indicates L- or D-stereochemistry;

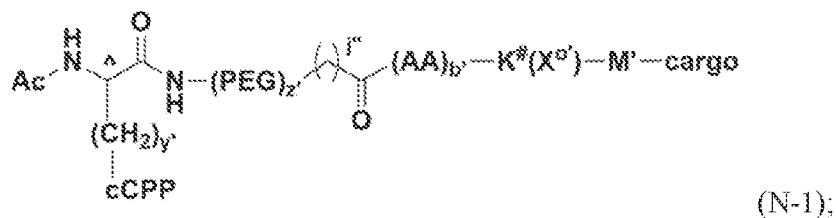
y' is an integer from 1 to 5;

M' comprises a bonding group; and

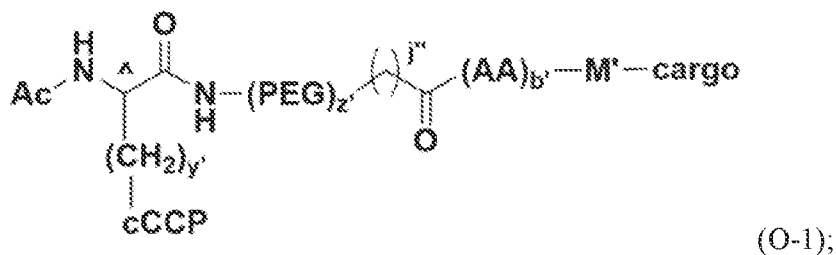
cargo is a peptide, an oligonucleotide, a small molecule, or any combinations thereof.

[0392] In embodiments, the compound Formula (AA-3) can have a Formula selected from (N-1), (O-1), (P-1), or (Q-1):

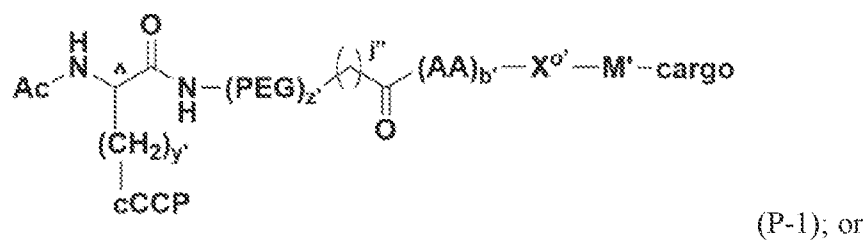
Formula (N-1):



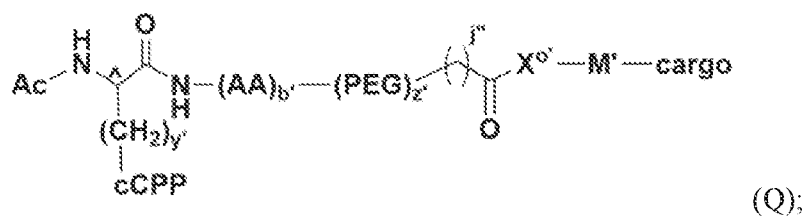
Formula (O-1):



Formula (P-1):



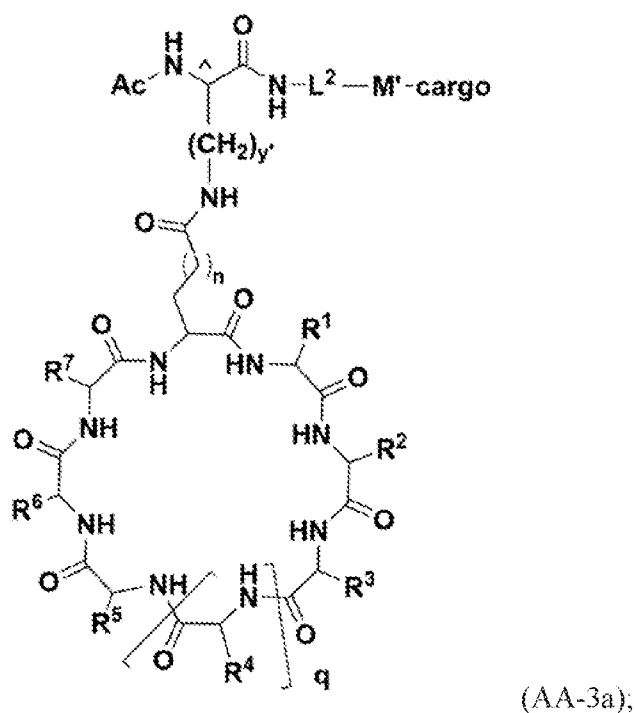
Formula (Q-1):



5 wherein Ac, cCPP, $\wedge$, y' , z' , j'' , $X^{o'}$, $K^\#$, (AA) b' , b' , M' , and cargo are as defined herein.

[0393] In embodiments, the EEV-cargo conjugate has the structure of formula (AA-3a):

Formula (AA-3a):



wherein:

10 Ac is an acetyl group ;

L^2 is a linker arm;

M' is a bonding group;

^ indicates L- or D-stereochemistry;

y' is an integer from 1-5,

R₁, R₂, and R₃ can each independently be H or an amino acid residue having a side chain comprising an aryl or heteroaryl group;

R₄, R₅, R₆ and R₇ are independently H or a side chain of an amino acid;

q is an integer from 1-4;

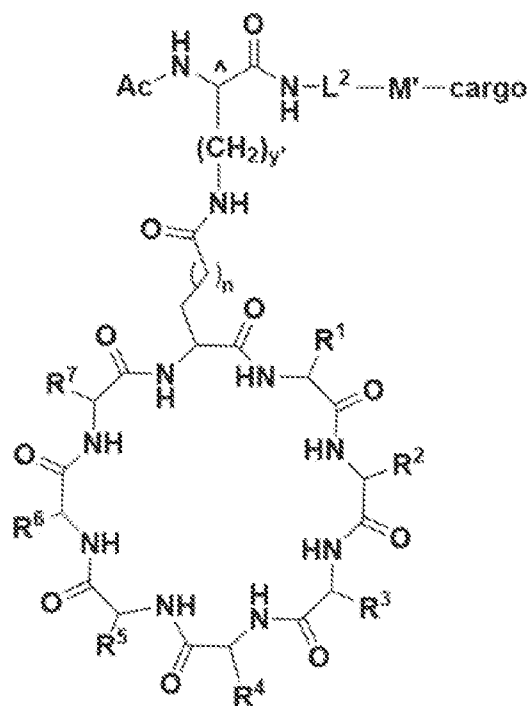
n is an integer from 1-4; and

cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

[0394] In embodiments, the EEV-cargo conjugate of Formula (A-3a) has a linker structure of Formula (N'), (O'), (P'), or (Q').

[0395] In embodiments, the EEV-cargo conjugate has the structure of formula (AA-3b):

Formula (AA-3b):



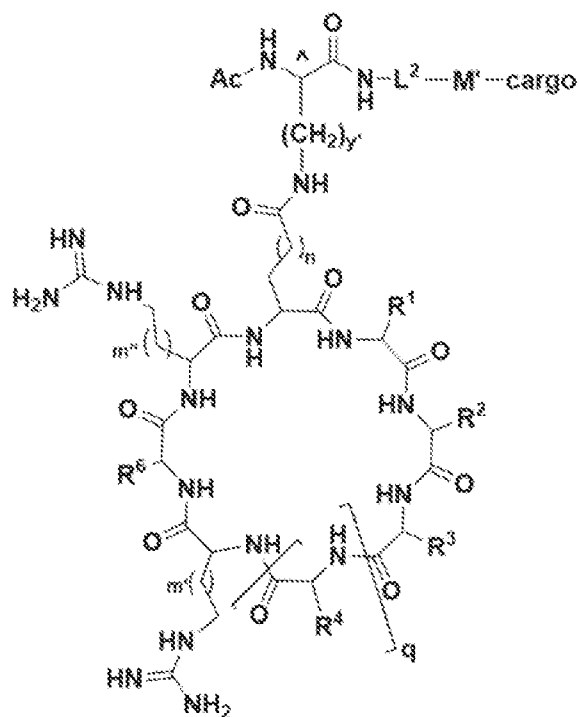
(A-3b);

wherein Ac, L², M', y', ^, n, R₁, R₂, R₃, R₄, R₅, R₆, and R₇ are as defined herein and cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

[0396] In embodiments, the EEV-cargo conjugate of Formula (AA-3b) has a linker structure of Formula (N'), (O'), (P'), or (Q').

[0397] In embodiments, the EEV-cargo conjugate has the structure of formula (AA-3c):

Formula (AA-3c):



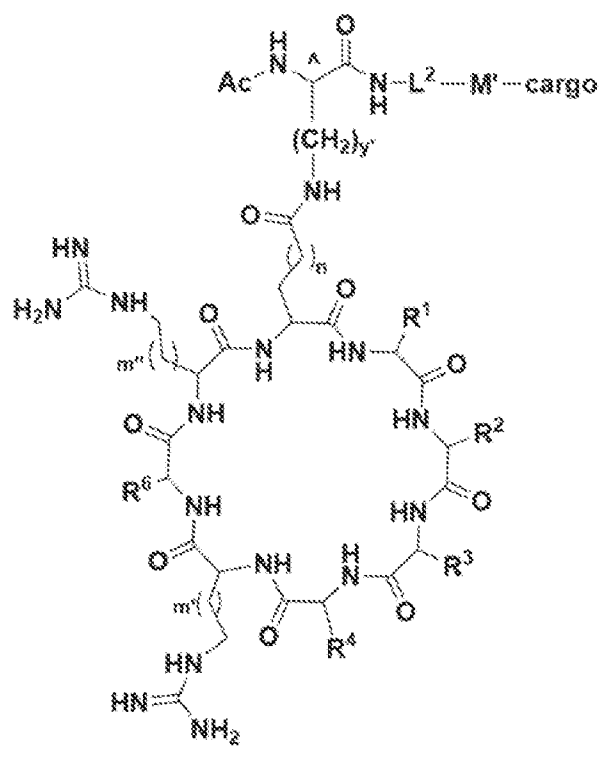
(AA-3c)

wherein Ac, L², M', y', ^, n, m', m'', q, R₁, R₂, R₃, R₄, and R₆ are as defined herein and cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

[0398] In embodiments, the EEV-cargo conjugate of Formula (AA-3c) has a linker structure of Formula (N'), (O'), (P'), or (Q').

[0399] In embodiments, the EEV-cargo conjugate has the structure of formula (AA-3d):

Formula (AA-3d):

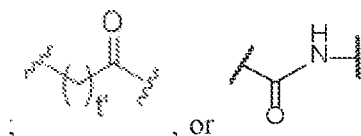
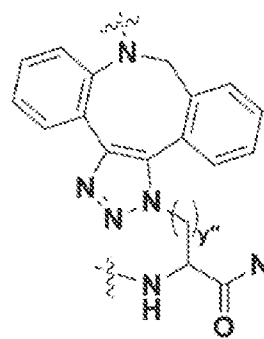
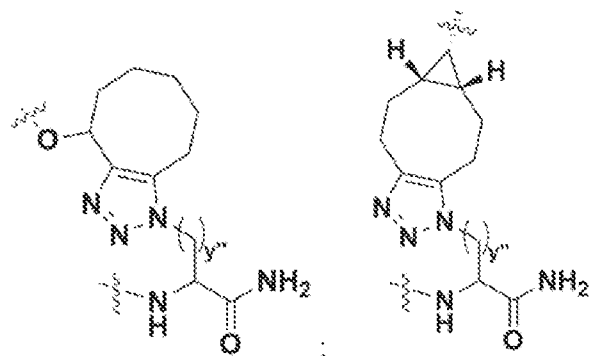


wherein Ac, L^2 , M' , y' , $\wedge$, n , m' , m'' , R_1 , R_2 , R_3 , R_4 , and R_6 are as defined herein and cargo is a peptide, oligonucleotide, a small molecule, or a combination thereof.

- 5 [0400] In embodiments, the EEV-cargo conjugate of Formula (AA-3d) has a linker structure of Formula (N'), (O'), (P'), or (Q').

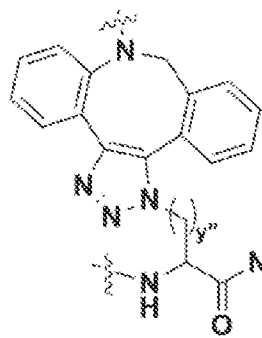
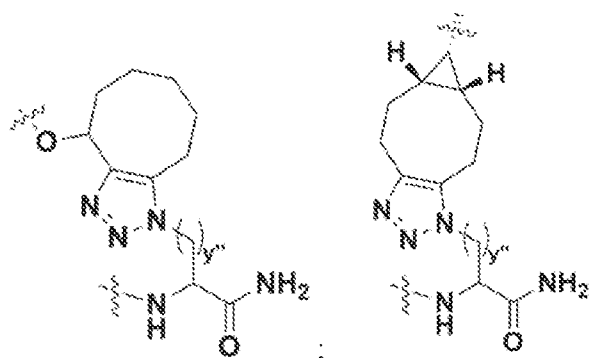
[0401] In embodiments, the cargo is an oligonucleotide. In embodiments, the oligonucleotide is an antisense oligonucleotide. In embodiments, the oligonucleotide is a phosphorodiamidate morpholino oligonucleotide (PMO).

[0402] M' can comprises: -NH-, -C(O)-, -O-,

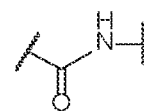
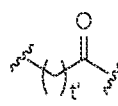
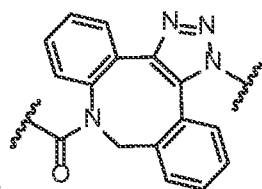


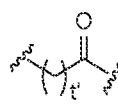
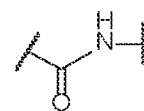
is 0 to 10.

[0403] M' can comprise: -NH-, -C(O)-, -O-,



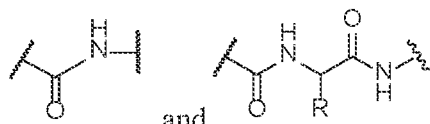
; wherein y'' is an integer from 1 to 4.

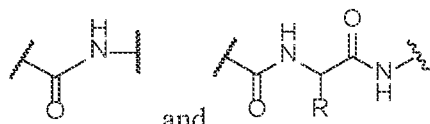
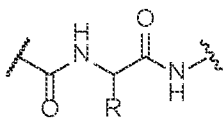


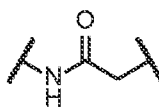
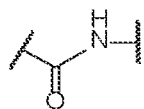
[0404] In embodiments, M' comprises , or , wherein t' is 0 to 10.

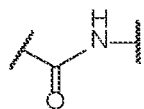
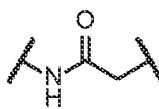
[0405] M' can comprise -NH-. M' can comprise -C(O)-. M' can comprise -O-.

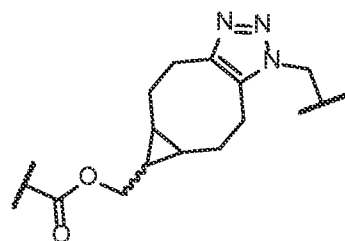
[0406] M' can comprise a structure selected from:



5 , and , wherein R is alkyl, alkenyl, alkynyl, carbocyclyl, or heterocyclyl.



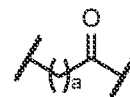
[0407] M' comprise a structure selected from: , , or



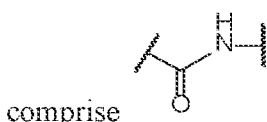
. M' can comprise



. M' can comprise

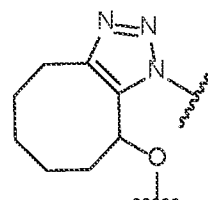
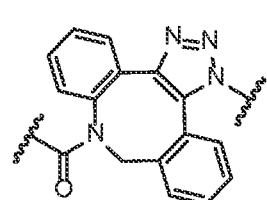


. M' can

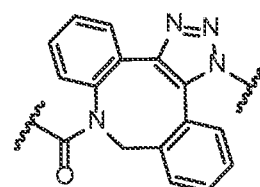


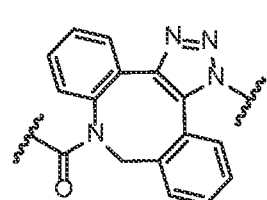
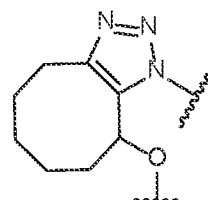
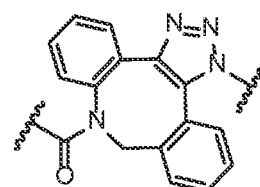
comprise

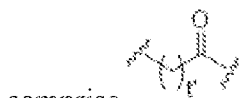
10 [0408] M' can comprise -C(O)-. M' can comprise -NH-. M' can comprise -O-.

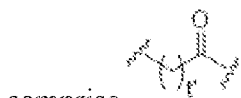


or

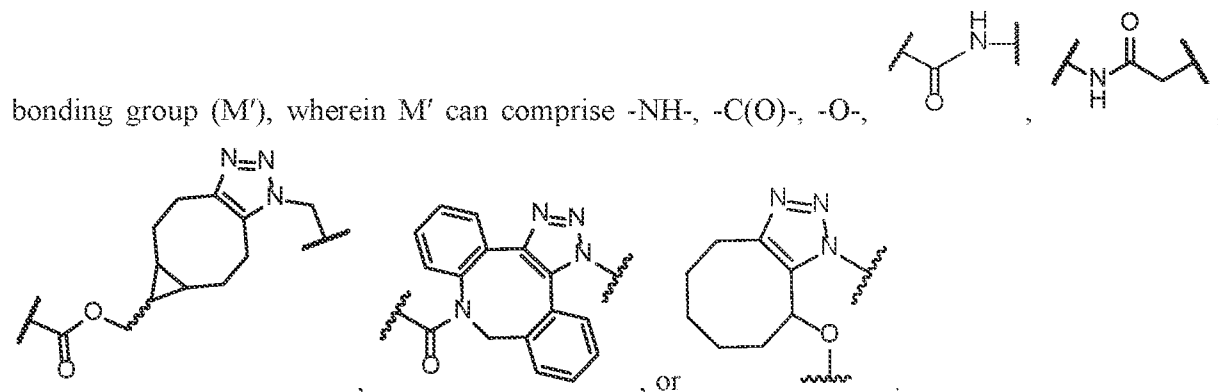


[0409] M' can comprise , , or . M' can



comprise , wherein t' is 0 to 10. M' can comprise . M' can be -O-. M' can be -NH-. M' can be -C(O)-,

[0410] Provided herein is a compound comprising an EEV conjugated to a cargo through a



5 [0411] The linker can be covalently bound to a cargo at any suitable location on the cargo. The linker can be covalently bound to the 3' end or 5' end of an oligonucleotide cargo. The linker can be covalently bound to the backbone of a cargo.

[0412] M' forms a bond to the free secondary amine of the morpholino ring of a terminal nucleotide of a PMO cargo. In embodiments, M' forms a bond to the free secondary amine of the morpholino ring of the 3' terminal nucleotide of a PMO. In embodiments, M' forms a bond to the free hydroxyl of the of the 5' terminal nucleotide of an PMO.

10

[0413] In embodiments, the EEV-cargo conjugate can be selected from:

Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-K(Ac-Bip)-M'-cargo
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-K(N3)-M'-cargo
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-M'-cargo
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-M'-cargo
Ac-KKK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RK-Nal-M'-cargo
Ac-KK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-KRK-Nal-M'-cargo
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-KKKRK-PEG ₁₂ -Nal-M'-cargo
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-M'-cargo
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-M'-cargo
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG12-kkkrk(Ac-nal)-M'-cargo

Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkr-nal-M'-cargo

[0414] In embodiments, the EEV-cargo conjugate can be selected from:

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -Nal-M'-cargo
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Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRGQ])-PEG ₁₂ -Nal-M'-cargo
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[0415] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Nal-PEG ₂ -Nal-M'-cargo
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[0416] In embodiments, the EEV-cargo conjugate is selected from:

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -dNal-M'-cargo

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -Nal-M'-cargo
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Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fffrrrrQ])-PEG ₁₂ -dNal-M'-cargo

[0417] In embodiments, the EEV-cargo conjugate is selected from:

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -dNal-M'-cargo

10 [0418] In embodiments, the EEV-cargo conjugate is selected from:

Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -dNal-M'-cargo

[0419] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-M'-cargo
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Ac-KKKRK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-M'-cargo
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Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-M'-cargo

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-M'-cargo
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Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-M'-cargo

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-M'-cargo

[0420] In embodiments, the EEV-cargo conjugate is selected from:

<i>cyclo</i> [FGFGRGRQ]-Lys(Bip-KRKKKP-Ac)-M'-cargo

<i>cyclo</i> [FGFGRGRQ]-Lys(PKKKRK-Bip-Ac)-M'-cargo

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[0421] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-M'-cargo

[0422] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG ₁₂ -Nal-M'-cargo
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5 [0423] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-M'-cargo

[0424] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG ₁₂ -Nal-M'-cargo

[0425] In embodiments, the EEV-cargo conjugate is selected from:

Ac-RBRRBR-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRRRRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRrRrQ])-PEG ₁₂ -Nal-M'-cargo and

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[0426] In embodiments, the EEV-cargo conjugate is selected from:

Ac-rbrrbr-PEG ₂ -K(cyclo[fGfGrGrQ])-PEG ₁₂ -Nal-M'-cargo
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[0427] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRGQ])-PEG ₁₂ -Nal-M'-cargo

15 [0428] In embodiments, the EEV is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo

Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-M'-cargo

[0429] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-K(cyclo[fFRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FAFARARQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FGFGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG ₁₂ -Nal-M'-cargo

[0430] In embodiments, the EEV-cargo conjugate is:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG ₁₂ -Nal-M'-cargo
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5 [0431] In embodiments, the EEV-cargo conjugate is selected from:

Ac-RBRRBR-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRRRRQ])-PEG ₁₂ -Nal-M'-cargo
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRrRrQ])-PEG ₁₂ -Nal-M'-cargo and
Ac-rbrrbr-PEG ₂ -K(cyclo[fGfGrGrQ])-PEG ₁₂ -Nal-M'-cargo

[0432] In embodiments, the EEV-cargo conjugate is selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG ₁₂ -dNal-M'-cargo
Ac-pkkkrkv-PEG ₂ -k(cyclo[fffrrrQ])-PEG ₁₂ -dNal-M'-cargo
Ac-pkkkrkv-PEG ₂ -k(cyclo[FGFRRRRQ])-PEG ₁₂ -dNal-M'-cargo and
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -dNal-M'-cargo

[0433] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-Bip-M'-cargo

[0434] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG₂-K(*cyclo*[FGFGRGRQ])-PEG₁₂-C₆-M'-cargo.

[0435] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -F-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -W-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Y-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -V-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -I-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -L-M'-cargo and
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -H-M'-cargo

[0436] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG₂-K(*cyclo*[FGFGRGRQ])-G-M'-cargo.

5 [0437] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG₂-K(*cyclo*[FGFGRGRQ])-PEG₁₂-Nal-PEG₂-Nal-M''-cargo.

[0438] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-M'-cargo
Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-M'-cargo
Ac-KKK-PEG ₂ -K(<i>cyclo</i> [Ff-Nal-GrGrQ])-PEG ₁₂ -RK-Nal-M'-cargo
Ac-KK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKK-Nal-M'-cargo
Ac-K-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-M'-cargo and
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-M'-cargo

[0439] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -rk-nal-M'-cargo
Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -rk-nal-M'-cargo
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkr-nal-M'-cargo and
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkr-nal-M'-cargo

[0440] In embodiments, the EEV-cargo conjugate is selected from

Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-M'-cargo and
Ac-PKKKRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-M'-cargo

10 [0441] In embodiments, the EEV-cargo conjugate is selected from

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-M'-cargo and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-M'-cargo

[0442] In embodiments, the EEV-cargo conjugate is selected from

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-M'-cargo and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-M'-cargo

[0443] In embodiments, the EEV-cargo conjugate is

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-M'-cargo

[0444] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKK-PEG ₂ -K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-M'-cargo
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-M'-cargo and
Ac-K(cyclo[FGFGRGRQ])-KKKRK-PEG ₁₂ -Nal-M'-cargo

[0445] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Nal)-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Bip)-M'-cargo and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkrk(Ac-nal)-M'-cargo

5 [0446] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-W)-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Y)-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-I)-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-L)-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-H)-M'-cargo and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-F)-M'-cargo

[0447] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BHRHR-M'-cargo and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRHRH-M'-cargo

[0448] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RFQILY-PEG ₂ -M'-cargo

[0449] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -R-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -F-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RRF-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RFR-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RGRRG-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RFRRF-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RBRBR-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -KKKRK-PEG ₄ -M'-cargo and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo

[0450] In embodiments, the EEV-cargo conjugate is:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₂-RF-PEG₄-M'-cargo.

5 [0451] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₄ -RF-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -RF-PEG ₄ -M'-cargo and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -RF-PEG ₄ -M'-cargo

[0452] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo

[0453] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₂ -Bip-M'-cargo

[0454] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-M'-cargo
Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-M'-cargo and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-M'-cargo

[0455] In embodiments, the EEV-cargo conjugate is:

10 Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₈-R-Bip-M'-cargo.

[0456] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKG-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-RB-Bip-RB-M'-cargo.

[0457] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(KKKRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-KKKRK-PEG ₂ -K(RBRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo

Ac-RBRBR-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo and
Ac-KRK-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo

[0458] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-RF-PEG ₄ -M'-cargo and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-RF-PEG ₄ -M'-cargo

[0459] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-GRF-PEG ₄ -M'-cargo and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-BRF-PEG ₄ -M'-cargo

[0460] In embodiments, the EEV-cargo conjugate is:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-GGE-M'-cargo
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[0461] In embodiments, the EEV-cargo conjugate is:

5 Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-GRRRGRRG-M'-cargo.

[0462] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKV-PEG₂-K(cyclo[FfFGRGRQ])-PEG₂-RFQILYBRBRB-M'-cargo.

[0463] In embodiments, the EEV-cargo conjugate is:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₈-RF-M'-cargo.

10 [0464] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-M'-cargo and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-M'-cargo

[0465] In embodiments, the EEV-cargo conjugate is:

Ac-PKKKRKG-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-RB-Bip-RB-M'-cargo.

[0466] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKKR-K(Ac-Nal)-M'-cargo

15 Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKR-K(Ac-Nal)-M'-cargo and

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KR-K(Ac-Nal)-M'-cargo.

[0467] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKKR-K(Ac-dNal)-M'-cargo

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKR-K(Ac-dNal)-M'-cargo and

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KR-K(Ac-dNal)-M'-cargo.

[0468] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}K-N^{uMe}R-N^{uMe}K(Ac-Nal)-M'-cargo

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}R-N^{uMe}K(Ac-Nal)-M'-cargo and

5 Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}R-N^{uMe}K(Ac-Nal)-M'-cargo.

[0469] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}K-N^{uMe}R-N^{uMe}K(Ac-dNal)-M'-cargo

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}R-N^{uMe}K(Ac-dNal)-M'-cargo and

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}R-N^{uMe}K(Ac-dNal)-M'-cargo.

10 [0470] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKKR-M'-cargo

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKR-M'-cargo and

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KR-M'-cargo.

[0471] In embodiments, the EEV-cargo conjugate is selected from:

15 Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKKR-M'-cargo

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KKR-M'-cargo and

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-KR-M'-cargo.

[0472] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}K-N^{uMe}R-M'-cargo

20 Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}R-M'-cargo and

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}R-M'-cargo.

[0473] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}K-N^{uMe}R-M'-cargo

Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}K-N^{uMe}R-M'-cargo and

25 Ac-K(cyclo[FGFGRGRQ])-PEG₁₂-N^{uMe}K-N^{uMe}R-M'-cargo.

[0474] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKR-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KR-Nal-M'-cargo and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-M'-cargo

[0475] In embodiments, the EEV-cargo conjugate is selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{uMe} K-N ^{uMe} K-N ^{uMe} K-N ^{uMe} R-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{uMe} K-N ^{uMe} K-N ^{uMe} R-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{uMe} K-N ^{uMe} K-N ^{uMe} R-Nal-M'-cargo
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{uMe} K-N ^{uMe} R-Nal-M'-cargo and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{uMe} K-N ^{uMe} K-N ^{uMe} R-Nal-M'-cargo

[0476] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BHRHR-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRHRH-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-M'-cargo
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-M'-cargo and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-M'-cargo

[0477] In embodiments, the EEV-cargo conjugate is selected from:

cyclo[FGFGRGRQ]-K(Bip-KRKKKP-Ac)-M'-cargo and
cyclo[FGFGRGRQ]-K(PKKKRK-Bip-Ac)-M'-cargo

[0478] In embodiments, the EEV-cargo conjugate is selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -M'-cargo and
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -M'-cargo

5 [0479] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo

Ac-KKKRK-PEG2-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo
Ac-KKKRK-PEG4-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo
Ac-KKKRK-PEG8-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo
Ac-KKKRK-PEG2-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo
Ac-KRK-PEG2-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo
Ac-RK-PEG2-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo
Ac-RF-PEG2-K(cyclo[FfFGRGRQ])-PEG8-RF-M'-cargo

[0480] In embodiments, the EEV-cargo conjugate is selected from:

Ac-pkkkrkv-PEG₂-k(cyclo[fGfrrrrQ])-PEG₁₂-dNal-M'-cargo

Ac-pkkkrkv-PEG₂-k(cyclo[ffrrrrQ])-PEG₁₂-dNal-M'-cargo and

Ac-pkkkrkv-PEG₂-k(cyclo[fGfGrGrQ])-PEG₁₂-dNal-M'-cargo.

[0481] In embodiments, the EEV-cargo conjugate is:

Ac-rbrrrbr-PEG₂-K(cyclo[fGfGrGrQ])-PEG₁₂-Nal-M'-cargo.

[0482] In embodiments, the EEV-cargo conjugate is:

5 Ac-pkkkrkv-PEG₂-k(cyclo[fGfrrrrQ])-PEG₁₂-Nal-M'-cargo.

[0483] In embodiments, the EEV-cargo conjugate is selected from:

cyclo[FGFGRGRQ]-K(Bip-KRKKKP-Ac)-M'-cargo or

cyclo[FGFGRGRQ]-K(PKKKRK-Bip-Ac)-M'-cargo.

[0484] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KRK-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-KRK-PEG ₂ -K(KRKIL-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-KRKIL-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-KRK-PEG ₂ -K(KRKF-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-KRKF-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo

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[0485] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-KKKRK-PEG ₂ -K(RBRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-RBRRBR-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo

[0486] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(RFRRF-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-RFRRF-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo

[0487] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(RIRRI-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo
Ac-RIRRI-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -M'-cargo

5 [0488] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -FGF-K(cyclo[RGRGRGRQ])-PEG ₁₂ -M'-cargo

[0489] In embodiments, the EEV-cargo conjugate is selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[RGRGRGRQ])-PEG ₁₂ -dNal-M'-cargo
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[0490] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -R-M'-cargo

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[0491] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo

[0492] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo

15 [0493] In embodiments, the EEV-cargo conjugate is selected from:

Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo
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[0494] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKFRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo
Ac-KFK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-M'-cargo

[0495] In embodiments, the EEV-cargo conjugate is selected from:

Ac-RF-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KKFRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo

[0496] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKFRK-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo

[0497] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGRGFGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FRFRFRFQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFLRLRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FGFVRVRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo

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[0498] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FffGRGRQ])-PEG ₂ -KRR-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FffGRGRQ])-PEG ₂ -KKRR-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FffGRGRQ])-PEG ₂ -KKKRR-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FffGRGRQ])-PEG ₂ -KRK-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FffGRGRQ])-PEG ₂ -KKRK-PEG ₄ -M'-cargo
Ac-KKKRK-PEG ₂ -K(cyclo[FffGRGRQ])-PEG ₂ -KKKRK-PEG ₄ -M'-cargo

[0499] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo
Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -M'-cargo

10 [0500] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo
Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo
Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -M'-cargo

[0501] In embodiments, the EEV-cargo conjugate is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo
Ac-KKKRK-PEG ₄ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo
Ac-KKKRK-PEG ₈ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo

[0502] In embodiments, the EEV is selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo
Ac-KRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo
Ac-RK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo
Ac-RF-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-M'-cargo

5 Cargo

[0503] The cargo conjugates provided herein comprise a delivery construct and a therapeutic moiety or therapeutic agent. The therapeutic moiety may be any suitable therapeutic moiety for treating a disease. As used herein, the terms “therapeutic moiety” or “therapeutic agent” refer to any molecule (e.g., polypeptide, small molecule, oligonucleotide, gene editing machinery, or combination thereof) that is designed to have and/or has prophylactic or other biological activity.

In embodiments, the therapeutic moiety selectively binds to a target molecule associated with a disease. In embodiments, the target molecule is a macromolecule implicated in a disease or pathology. In embodiments, the target molecule is a polypeptide or protein. In embodiments, the target molecule is an oligonucleotide. In embodiments, the oligonucleotide target comprises DNA.

In embodiments, the oligonucleotide target comprises genomic DNA. In embodiments, the oligonucleotide target comprises RNA. In embodiments, the oligonucleotide target comprises mRNA. In embodiments, the target molecule is associated with a disease. In embodiments, selective binding of the therapeutic moiety with the target molecule is useful for the treatment of a disease, pathology or other abnormal state or condition. In embodiments, selective binding of the therapeutic moiety to the target molecule upregulates expression or activity of the target molecule. In embodiments, selective binding of the therapeutic moiety to the target molecule downregulates expression or activity of the target molecule.

[0504] In embodiments, the therapeutic moiety comprises a therapeutic oligonucleotide. In embodiments, the therapeutic moiety comprises a polypeptide. In embodiments, the therapeutic

moiety comprises a small molecule. In embodiments, the therapeutic moiety includes one or more components of gene editing machinery.

Therapeutic Oligonucleotides

[0505] In embodiments, the therapeutic moiety comprises a therapeutic oligonucleotide. In
5 embodiments, the therapeutic oligonucleotide comprises an antisense oligonucleotide (ASO). In
embodiments, the therapeutic oligonucleotide comprises siRNA, RNAi, microRNA, antagomir,
an aptamer, a ribozyme, an immunostimulatory oligonucleotide, a decoy oligonucleotide, a
supermir, a miRNA mimic, a miRNA inhibitor, or a combination thereof (See, for example, Chery,
J., “RNA therapeutics: RNAi and antisense mechanisms and clinical applications,” Postdoc J, July
10 2016, 4(7):35-50, and Zhu, et al., “RNA-based therapeutics: an overview and prospectus, Cell
Death & Disease, 23 July 2022, 12(644) (doi: 10.1038/s41419-022-05075-2).

[0506] In embodiments, therapeutic oligonucleotides are provided that include from about 5 to
about 100 nucleic acids. In embodiments, the therapeutic oligonucleotide is from about 5 to about
50, about 8 to about 40, about 10 to about 30, about 15 to about 30, or about 20 to about 30
15 nucleotides in length. In embodiments, the therapeutic oligonucleotide includes one or more
modified nucleosides, one or more modified internucleoside linkages, one or more conjugate
groups, or combinations thereof.

Antisense Oligonucleotides (ASO)

[0507] In embodiments, the therapeutic oligonucleotide is an antisense oligonucleotide (ASO)
20 directed to a target gene or a target transcript associated with a disease. The ASO may be directed
to and bind to a target nucleotide sequence located within a target gene or a target transcript. In
embodiments, the target nucleotide sequence is within a target gene and/or target transcript
associated with a disease.

[0508] The term “antisense oligonucleotide” refers to an oligonucleotide that is at least partially
25 complementary to a target sequence within a target polynucleotide. An antisense oligonucleotide
(ASO) is a single stranded molecule that contains DNA, RNA, or combinations or modifications
thereof that are at least partially complementary to a chosen sequence, e.g., a target nucleotide
sequence within a target gene or target transcript.

[0509] An ASO may modulate one or more aspects of gene expression and/or protein function via
30 hybridization of the ASO with a target nucleotide sequence. The ASO may modulate one or more
aspects of gene expression and/or protein function through various mechanisms. For example,

hybridization of an ASO to a target nucleotide sequence of a target transcript may modulate splicing of a target transcript such as, for example, via exon skipping; exon inclusion; alternative splicing; prevent polyadenylation of the target transcript; increase the target transcript stability; induce target transcript degradation; prevent translation of the target transcript; or any combination thereof. ASOs have been demonstrated to be effective and targeted inhibitors of protein synthesis, and, consequently, can be used to modulate gene expression of a targeted gene.

[0510] In embodiments, the ASO hybridizes with a target sequence of a target gene or target transcript having a sequence from about 5 to about 50 nucleotides in length, which can also be referred to as the length of the ASO. In embodiments, the ASO is from about 5 to about 50, about 8 to about 40, about 10 to about 30, about 15 to about 30, or about 20 to about 30 nucleotides in length. In embodiments, the ASO is at least about 5, about 6, about 7, about 8, about 9, about 10, about 11, about 12, about 13, about 14, or about 15, and up to about 16, about 17, about 18, about 19, about 20, about 21, about 22, about 23, about 24, about 25, about 26, about 27, about 28, about 29, about 30, about 31, about 32, about 33, about 34, about 35, about 36, about 37, about 38, about 39, about 40, about 41, about 42, about 43, about 44, about 45, about 46, about 47, about 48, about 49, or about 50 nucleotides in length. In embodiments, the ASO is about 15 nucleotides in length. In embodiments, the ASO is about 16 nucleotides in length. In embodiments, the ASO is about 17 nucleotides in length. In embodiments, the ASO is about 18 nucleotides in length. In embodiments, the ASO is about 19 nucleotides in length. In embodiments, the ASO is about 20 nucleotides in length. In embodiments, the ASO is about 21 nucleotides in length. In embodiments, the ASO is about 22 nucleotides in length. In embodiments, the ASO is about 23 nucleotides in length. In embodiments, the ASO is about 24 nucleotides in length. In embodiments, the ASO is about 25 nucleotides in length. In embodiments, the ASO is about 26 nucleotides in length. In embodiments, the ASO is about 27 nucleotides in length. In embodiments, the ASO is about 28 nucleotides in length. In embodiments, the ASO is about 29 nucleotides in length. In embodiments, the ASO is about 30 nucleotides in length.

[0511] In embodiments, the ASO may be less than about 100 percent complementary to a target nucleotide sequence. As used herein, the term “percent complementarity” refers to the number of nucleobases of an ASO that have nucleobase complementarity with a corresponding nucleobase of target nucleotide sequence by the total length (number of nucleobases) of the ASO. One skilled in the art recognizes that the inclusion of mismatches is possible without eliminating the activity

of the antisense compound. In embodiments, the ASOs contain no more than about 15%, no more than about 10%, no more than 5%, or no mismatches. In embodiments, the ASOs are at least about 80%, at least about 85%, at least about 90%, at least about 95%, at least about 96%, at least about 97%, at least about 98%, at least about 99%, about 100%, or 100% complementary to a target nucleic acid. Percent complementarity of an oligonucleotide is calculated by dividing the number of complementary nucleobases by the total number of nucleobases of the oligonucleotide. Percent complementarity of a region of an oligonucleotide is calculated by dividing the number of complementary nucleobases in the region by the total number of nucleobases region.

[0512] In embodiments, incorporation of nucleotide affinity modifications allows for a greater number of mismatches compared to an unmodified compound. Similarly, certain oligonucleotide sequences may be more tolerant to mismatches than other oligonucleotide sequences. One of ordinary skill in the art is capable of determining an appropriate number of mismatches between an ASO and a target nucleotide sequence, such as by determining melting temperature (T_m). T_m or change in T_m (ΔT_m) can be calculated by techniques that are familiar to one of ordinary skill in the art. For example, techniques described in Freier et al. (Nucleic Acids Research, 1997, 25, 22: 4429-4443) allow one of ordinary skill in the art to evaluate nucleotide modifications for their ability to increase the melting temperature of an RNA:DNA duplex.

Therapeutic oligonucleotides design

[0513] Design of a therapeutic oligonucleotides (e.g., an ASO) will depend upon the target gene. Targeting a therapeutic oligonucleotide to a particular target nucleotide sequence can be a multistep process. The process usually begins with the identification of gene of interest. The transcript of the gene of interest is analyzed and a target nucleotide sequence is identified. In embodiments, the target gene is a gene associated with a disease.

[0514] One of skill in the art will be able to design, synthesize, and screen therapeutic oligonucleotides of different nucleobase sequences to identify a sequence that results in antisense activity. For example, a therapeutic oligonucleotide can be designed that inhibits expression of a target gene. Methods for designing, synthesizing, and screening therapeutic nucleotides for antisense activity against a preselected target nucleic acid and/or target gene can be found, for example in "Antisense Drug Technology, Principles, Strategies, and Applications" Edited by Stanley T. Crooke, CRC Press, Boca Raton, Florida, which is incorporated by reference in its entirety for any purpose.

[0515] The efficacy of a therapeutic oligonucleotide (e.g., an ASO) may be assessed by evaluating the antisense activity effected by their administration. As used herein, the term "antisense activity" refers to any detectable and/or measurable activity attributable to the hybridization of a therapeutic oligonucleotide to its target nucleotide sequence. Such detection and/or measuring may be direct or indirect. In embodiments, antisense activity is assessed by detecting and or measuring the amount of target protein in a cell or population of cells before and after administration of the therapeutic oligonucleotide to the cell or population of cells. In embodiments, antisense activity is assessed by detecting and/or measuring the amount of target transcript in a cell or population of cells.

10 *Therapeutic Oligonucleotides Structure*

[0516] Therapeutic oligonucleotides comprise nucleosides linked through internucleoside linkages. Nucleosides include a pentose sugar (e.g., ribose or deoxyribose) and a nitrogenous base (nucleobase or simply base) covalently attached to sugar. The naturally occurring (traditional) bases found in DNA and/or RNA are adenine (A), guanine (G), thymine (T), cytosine (C), and uracil (U). The naturally occurring (traditional) sugars found in DNA and/or RNA deoxyribose (DNA) and ribose (RNA). A naturally occurring (traditional) nucleoside linkage is a phosphodiester bond. In embodiments, the therapeutic oligonucleotides may have all natural sugars, natural bases, and natural internucleoside linkages.

[0517] Chemically modified nucleosides are routinely used for incorporation into therapeutic oligonucleotides to enhance one or more properties, such as nuclease resistance, pharmacokinetics, or affinity for a target gene or target transcript. Non-limiting examples of nucleosides are provided in Khvorova et al. Nature Biotechnology (2017) 35: 238-248, which is incorporated by reference herein in its entirety. In embodiments, a therapeutic oligonucleotide has one or more modified nucleosides. In embodiments, a therapeutic oligonucleotide has one or more modified sugars. In 25 embodiments, a therapeutic oligonucleotide has one or more modified bases. In embodiments, a therapeutic oligonucleotide has one or more modified internucleoside linkages.

[0518] In general, a nucleobase is any group that contains one or more atoms or groups of atoms capable of hydrogen bonding to a base of another nucleic acid. In addition to natural nucleobases, many modified nucleobases or nucleobase mimetics known to those skilled in the art are amenable with the compounds described herein. The terms modified nucleobase and nucleobase mimetic can 30 overlap, but generally a modified nucleobase refers to a nucleobase that is fairly similar in structure

to the parent nucleobase, such as for example a 7-deaza purine, a 5-methyl cytosine, or a G-clamp, whereas a nucleobase mimetic generally includes more complicated structures, such as for example a tricyclic phenoxazine nucleobase mimetic. Methods for preparation of the above noted modified nucleobases are well known to those skilled in the art.

5 [0519] In embodiments, a therapeutic oligonucleotide includes one or more nucleosides having a modified sugar moiety. In embodiments, the furanosyl sugar ring of a natural nucleoside can be modified. A furanosyl sugar ring may be modified in any suitable manner, including, but not limited to, addition of a substituent group, bridging of two non-geminal ring atoms to form a bicyclic nucleic acid (BNA) and substitution of an atom or group such as -S-, -N(R)- or -C(R1)(R2)
10 for the ring oxygen at the 4'-position. Modified sugar moieties are well known and can be used to alter, typically increase, the affinity of the antisense compound for its target and/or increase nuclease resistance. A representative list of modified sugars includes, but is not limited to, non-bicyclic substituted sugars, especially non-bicyclic 2'-substituted sugars having a 2'-F, 2'-OCH₃ or a 2'-O(CH₂)₂-OCH₃ substituent group; and 4'-thio modified sugars. Sugars can also be replaced
15 with a sugar mimetic group, for example, a morpholino ring, a methylenemorpholine ring, among others.

[0520] In embodiments, a therapeutic oligonucleotide may include one or more bicyclic modified sugars (BNA's), such as, for example, LNA (4'-(CH₂)-O-2' bridge), 2'-thio-LNA (4'-(CH₂)-S-2' bridge), 2'-amino-LNA (4'-(CH₂)-NR-2' bridge), ENA (4'-(CH₂)₂-O-2' bridge), 4'-(CH₂)₃-2' bridged BNA, 4'-(CH₂CH(CH₃))-2' bridged BNA" cEt (4'-(CH(CH₃))-O-2' bridge), and cMOE
20 BNAs (4'-(CH(CH₂OCH₃))-O-2' bridge).

[0521] In embodiments, a therapeutic oligonucleotide may include one or more locked nucleic acids" (LNAs) in which the 2'-hydroxyl group of the ribosyl sugar ring is linked to the 4' carbon atom of the sugar ring thereby forming a 2'-C,4'-C-oxymethylene linkage to form the bicyclic sugar
25 moiety. The synthesis and preparation of the LNA monomers adenine, cytosine, guanine, 5-methyl-cytosine, thymine and uracil, along with their oligomerization, and nucleic acid recognition properties have been described (Koshkin et al., Tetrahedron, 1998, 54, 3607-3630). LNAs and preparation thereof are also described in WO 98/39352 and WO 99/14226.

[0522] Internucleoside linking groups link the nucleosides or otherwise modified monomer units
30 of an oligonucleotide together. The two main classes of internucleoside linking groups are defined by the presence or absence of a phosphorus atom. Representative phosphorus containing

internucleoside linkages include, but are not limited to, phosphodiester, phosphotriester, methylphosphonates, phosphoramidate, phosphorodiamidate, and phosphorothioates. Representative non-phosphorus containing internucleoside linking groups include, but are not limited to, methylenemethylimino (-CH₂-N(CH₃)-O-CH₂-), thiodiester (-O-C(O)-S-),
5 thionocarbamate (-O-C(O)(NH)-S-); siloxane (-O-Si(H)₂-O-); and N,N'-dimethylhydrazine (-CH₂-N(CH₃)-N(CH₃)-). Therapeutic oligonucleotides having non-phosphorus internucleoside linking groups are referred to as oligonucleosides. Modified internucleoside linkages, compared to natural phosphodiester linkages, can be used to alter, typically increase, nuclease resistance of the therapeutic oligonucleotides. Internucleoside linkages having a chiral atom can be prepared
10 racemic, chiral, or as a mixture. Representative chiral internucleoside linkages include, but are not limited to, alkylphosphonates and phosphorothioates. Methods of preparation of phosphorous-containing and non-phosphorous-containing linkages are well known to those skilled in the art.

[0523] In embodiments, a phosphate group can be linked to the 2', 3' or 5' (or 6', for a 6 membered ring, such as a methylenemorpholine ring) hydroxyl moiety of the sugar (or sugar mimetic). In
15 forming oligonucleotides, the phosphate groups covalently link adjacent nucleosides to one another to form a linear polymeric compound. Within oligonucleotides, the phosphate groups are commonly referred to as forming the internucleoside backbone of the oligonucleotide. The normal linkage or backbone of RNA and DNA is a 3' to 5' phosphodiester linkage. In embodiments, the oligonucleotide is a phosphorodiamidate morpholino oligomer (PMO) comprising a backbone of
20 methylenemorpholine rings linked through phosphorodiamidate internucleoside linkages.

[0524] PMOs are uncharged nucleic acid analogs bind to target nucleic acid through base pairing. PMOs that bind to mRNA may block interaction of proteins to the mRNA through steric blockade (See, e.g., Nan and Zhang, *Front. Microbiol.* 20 April 2019 (doi.org/10.3389/fmicb.2018.00750)). As uncharged, or net neutral charged, oligonucleotides, PMOs are particularly effective for
25 intracellular delivery with a delivery construct.

[0525] The therapeutic oligonucleotides may contain one or more asymmetric centers and thus give rise to enantiomers, diastereomers, and other stereoisomeric configurations that may be defined, in terms of absolute stereochemistry, as (R) or (S); α or β ; or as (D) or (L). Included in the antisense compounds provided herein are all such possible isomers, as well as their racemic
30 and optically pure forms.

[0526] In embodiments, therapeutic oligonucleotides are modified by covalent attachment of one or more conjugate groups. In general, conjugate groups modify one or more properties of the attached therapeutic oligonucleotides including but not limited to pharmacodynamic, pharmacokinetic, binding, absorption, cellular distribution, cellular uptake, charge and clearance.

5 Conjugate groups are routinely used in the chemical arts and are linked directly or via a bonding group to a parent compound such as a therapeutic oligonucleotides. Conjugate groups include without limitation, intercalators, reporter molecules, polyamines, polyamides, polyethylene glycols, thioethers, polyethers, cholesterol, thiocholesterols, cholic acid moieties, folate, lipids, phospholipids, biotin, phenazine, phenanthridine, anthraquinone, adamantane, acridine, fluoresceins, rhodamines, coumarins and dyes. In embodiments, the conjugate group is a
10 polyethylene glycol (PEG), and the PEG is conjugated to either the therapeutic oligonucleotide, a linker, an EP, or the cyclic peptide.

Gene-Editing Machinery

[0527] In embodiments, the therapeutic moiety comprises one or more component of gene-editing
15 machinery. As used herein, “gene-editing machinery” refers to protein, nucleic acids, or combinations thereof, which may be used to edit a genome. Non-limiting examples of gene-editing machinery include guide RNAs (gRNAs), nucleases, nuclease inhibitors, and combinations and complexes thereof.

[0528] The gene editing machinery may be used to repair a mutated gene or to introduce a mutation
20 into a gene. The gene may be a gene associated with a disease.

[0529] In embodiments, a linker conjugates the delivery construct to the one or more components of gene-editing machinery. Any linker described in this disclosure or that is known to a person of skill in the art may be utilized.

gRNA

25 [0530] In embodiments, the therapeutic moiety includes a guide RNA (gRNA). A gRNA targets a genomic loci in a prokaryotic or eukaryotic cell.

[0531] In embodiments, the gRNA is a single-molecule guide RNA (sgRNA). A sgRNA includes a spacer sequence and a scaffold sequence. A spacer sequence is a short nucleic acid sequence used to target a nuclease (e.g., a Cas9 nuclease) to a specific nucleotide region of interest (e.g., a
30 genomic DNA sequence to be cleaved). In embodiments, the spacer may be about 17-24 bases in length, such as about 20 bases in length.

[0532] In embodiments, the spacer targets a site that immediately precedes a 5' protospacer adjacent motif (PAM). The PAM sequence may be selected based on the desired nuclease. For example, the PAM sequence may be any one of the PAM sequences shown in **Table 2** below, wherein N refers to any nucleic acid, R refers to A or G, Y refers to C or T, W refers to A or T, and V refers to A or C or G.

Table 2. Nucleases and PAM sequences

PAM sequence (5' to 3')	Nuclease	Isolated from
NGG	SpCas9	Streptococcus pyogenes
NGRRT or NGRRN	SaCas9	Staphylococcus aureus
NNNGATT	NmeCas9	Neisseria meningitidis
NNNNRYAC	CjCas9	Campylobacter jejuni
NNAGAAW	StCas9	Streptococcus thermophiles
TTTV	LbCpfI	Lachnospiraceae bacterium
TTTV	AsCpfI	Acidaminococcus sp.

[0533] In embodiments, a spacer may target a sequence of a mammalian gene, such as a human gene. In embodiments, the spacer may target a mutant gene. In embodiments, the spacer may target a coding sequence. In embodiments, the spacer may target an exonic sequence. In embodiments, the spacer may target a polyadenylation site (PS). In embodiments, the spacer may target a sequence element of a PS. In embodiments, the spacer may target a polyadenylation signal (PAS), an intervening sequence (IS), a cleavage site (CS), a downstream element (DES), or a portion or combination thereof. In embodiments, a spacer may target a splicing element (SE) or a cis-splicing regulatory element (SRE).

[0534] The scaffold sequence is the sequence within the sgRNA that is responsible for nuclease (e.g., Cas9) binding. The scaffold sequence does not include the spacer/targeting sequence. In embodiments, the scaffold may be about 10 to about 150 nucleotides in length, or about 50 to about 100 nucleotides in length.

[0535] In embodiments, the gRNA is single guide RNA molecule comprising the spacer and the scaffold. In embodiments, the gRNA comprises two molecules that hybridize to form the gRNA. An example of a gRNA that includes two molecules is a gRNA comprising a crRNA and

tracrRNA. In embodiments, the gRNA or one or more components thereof may further include a poly(A) tail.

[0536] In embodiments, a compound that includes a CPP is conjugated to a nucleic acid that includes a gRNA or a component thereof. In embodiments, the nucleic acid includes about 1, about 2, about 3, about 4, about 5, about 6, about 7, about 8, about 9, about 10, about 11, about 12, about 13, about 14, about 15, about 16, about 17, about 18, about 19, or about 20 gRNAs or components thereof. In embodiments, the gRNAs recognize the same target. In embodiments, the gRNAs recognize different targets.

[0537] In embodiments, a compound that includes a CPP is conjugated to a nucleic acid designed to express the gRNA within a cell. The nucleic acid may include a promoter sequence to drive expression of the gRNA.

Nuclease

[0538] In embodiments, the therapeutic moiety includes a nuclease. In embodiments, the nuclease is a Type II, Type V-A, Type V-B, Type VC, Type V-U, Type VI-B nuclease. In embodiments, the nuclease is a transcription, activator-like effector nuclease (TALEN), a meganuclease, or a zinc-finger nuclease or a modified form or variant thereof. In embodiments, the nuclease is a Cas9, Cas12a (Cpf1), Cas12b, Cas12c, Tnp-B like, Cas13a (C2c2), Cas13b, or Cas14 nuclease or a modified form or variant thereof. For example, in embodiments, the nuclease is a Cas9 nuclease or a Cpf1 nuclease.

[0539] In embodiments, the delivery construct is conjugated to a nucleic acid encoding a nuclease. In embodiments, the nucleic acid encoding a nuclease includes a sequence encoding a promoter, wherein the promoter drives expression of the nuclease.

gRNA and Nuclease Combinations

[0540] In embodiments, the therapeutic moiety includes a ribonucleoprotein (RNP) that includes a gRNA and a nuclease. A RNP is a complex of a gRNA bound to a nuclease. The gRNA, the nuclease, or both may be covalently attached to a delivery construct to form a cargo conjugate having an RNP therapeutic moiety.

[0541] In embodiments, a composition that includes: (a) a cargo conjugate comprising a delivery construct conjugated to a gRNA and (b) a nuclease is delivered to a cell. In embodiments, a composition that includes: (a) a cargo conjugate comprising a delivery construct conjugated to a nuclease and (b) an gRNA is delivered to a cell. In embodiments, a composition that includes: (a)

a first cargo conjugate comprising a first delivery construct conjugated to a gRNA and (b) a second cargo conjugate comprising a second delivery construct conjugated to a nuclease is delivered to a cell. In embodiments, the first delivery construct and the second delivery construct are the same. In embodiments, the first delivery construct and the second delivery construct are different.

- 5 [0542] In embodiments, a cargo conjugate comprises a delivery construct conjugated to a nucleic acid encoding a gRNA and/or a nuclease. In embodiments, the nucleic acid encoding a nuclease and a gRNA includes a sequence encoding a promoter, wherein the promoter drives expression of the nuclease and the gRNA. In embodiments, the nucleic acid encoding a nuclease and a gRNA includes two promoters, wherein a first promoter controls expression of the nuclease and a second
10 promoter controls expression of the gRNA. In embodiments, the nucleic acid encoding a gRNA and a nuclease encodes from about 1 to about 20 gRNAs, or from about 1, about 2, about 3, about 4, about 5, about 6, about 7, about 8, about 9, about 10, about 11, about 12, about 13, about 14, about 15, about 16, about 17, about 18, or about 19, and up to about 20 gRNAs. In embodiments, the gRNAs recognize different targets. In embodiments, the gRNAs recognize the same target.

15 *Nuclease Inhibitors*

[0543] In embodiments, the therapeutic moiety includes a nuclease inhibitor. A limitation of gene editing is potential off-target editing. The delivery of a nuclease inhibitor may limit off-target editing. In embodiments, the nuclease inhibitor is a polypeptide, polynucleotide, or small molecule.

20 *Therapeutic polypeptides*

- [0544] In embodiments, the therapeutic moiety includes a therapeutic polypeptide. In embodiments, the therapeutic polypeptide includes a peptide inhibitor. In embodiments, the peptide inhibitor inhibits a protein associated with a disease. In embodiments, the therapeutic polypeptide includes a peptide replacement therapy to functionally replace an aberrantly expressed
25 protein associated with a disease.

[0545] In embodiments, the therapeutic moiety includes a protein or a fragment thereof. In embodiments, the therapeutic moiety includes an RNA binding protein or an RNA binding fragment thereof. In embodiments, the therapeutic moiety includes an enzyme. In embodiments, the therapeutic moiety includes an RNA-cleaving enzyme or an active fragment thereof.

Antibodies

[0546] In embodiments, the therapeutic moiety includes an antibody or an antigen-binding fragment. Antibodies and antigen-binding fragments can be derived from any suitable source, including human, mouse, camelid (e.g., camel, alpaca, llama), rat, ungulates, or non-human primates (e.g., monkey, rhesus macaque).

[0547] The term “antibody” includes intact polyclonal or monoclonal antibodies and antigen-binding fragments thereof. For example, a native immunoglobulin molecule includes two heavy chain polypeptides and two light chain polypeptides. Each of the heavy chain polypeptides associate with a light chain polypeptide by virtue of interchain disulfide bonds between the heavy and light chain polypeptides to form two heterodimeric proteins or polypeptides (i.e., a protein that includes two heterologous polypeptide chains). The two heterodimeric proteins then associate by virtue of additional interchain disulfide bonds between the heavy chain polypeptides to form an immunoglobulin protein or polypeptide.

[0548] In embodiments, the therapeutic moiety is an antigen-binding fragment that binds to a target protein associated with a disease. An antibody may modulate the activity of the target protein to which it binds. In embodiments, the therapeutic moiety is an antigen-binding fragment that binds to a target transcript of a protein (Ye et al., *PNAS* (2008), 105(1):82-87; and Jung et al., (*RNA* (2014), 20(6):805-814). In embodiments, an antigen-binding fragment that binds to a target protein includes 1, 2, 3, 4, 5, or all 6 CDRs of a variable heavy chain (VH) and/or a variable light chain (VL) sequence from an antibody that specifically binds to the target protein. In embodiments, the antigen binding fragment includes 1, 2, or 3 of the CDRs of a camelid single domain antibody such as the VHH region. In embodiments, the antigen-binding fragment that binds to a target protein is a portion of a full-length antibody, such as Fab, F(ab')₂, Fab', Fv fragments, minibodies, diabodies, single domain antibody (dAb), single-chain variable fragments (scFv), multispecific antibodies formed from antibody fragments, or any other modified configuration of the immunoglobulin molecule that includes an antigen-binding site or fragment of the required specificity.

[0549] In embodiments, the therapeutic moiety includes a bispecific antibody. Bispecific antibodies (BsAbs) are antibodies that can simultaneously bind two separate and unique antigens (or different epitopes of the same antigen). In embodiments, the therapeutic moiety includes a bispecific antibody that can simultaneously bind to a target protein associated with a disease and

another target protein. Non-limiting examples include scFv (single-chain variable fragment), BsDb (bispecific diabody), scBsDb (single-chain bispecific diabody), scBsTaFv (single-chain bispecific tandem variable domain), DNL-(Fab)₃ (dock-and-lock trivalent Fab), sdAb (single-domain antibody), and BssdAb (bispecific single-domain antibody).

5 [0550] BsAbs with an Fc region are useful for carrying out Fc mediated effector functions such as antibody-dependent cell-mediated cytotoxicity and complement-dependent cytotoxicity. They have the half-life of normal IgG. On the other hand, BsAbs without the Fc region (bispecific fragments) rely solely on their antigen-binding capacity for carrying out therapeutic activity. Due to their smaller size, these fragments have better solid-tumor penetration rates. BsAb fragments do
10 not require glycosylation, and they may be produced in bacterial cells. The size, valency, flexibility and half-life of BsAbs to suit the application.

[0551] In embodiments, the therapeutic moiety includes a “diabody.” The term diabody refers to a bispecific antigen-binding antibody fragment in which VH and VL domains are expressed in a single polypeptide chain using a linker that is too short to allow for pairing between the two
15 domains on the same chain, thereby forcing the domains to pair with complementary domains of another chain and creating two antigen-binding sites (see, e.g., Holliger et al., Proc. Natl. Acad. Sci. USA 90:6444-48 (1993) and Poljak et al., Structure 2:1121- 23 (1994)). Diabodies may be designed to bind to two distinct antigens and are bi-specific antigen binding constructs.

[0552] In embodiments, the therapeutic moiety includes a “nanobody” or a “single domain antibody” (which can also be referred to herein as sdAbs or VHH). Single domain antibody refers
20 to an antigen-binding fragment that includes a single monomeric variable antibody domain comprising one variable domain (VH) of a heavy-chain antibody. In embodiments, the variable chain is the VHH of a camelid single chain antibody.

[0553] In embodiments, the therapeutic moiety includes a minibody.

25 [0554] In embodiments, the therapeutic moiety is an antibody mimetic. Antibody mimetics are compounds that, like antibodies, can specifically bind antigens, but that are not structurally related to antibodies. They are usually artificial peptides or proteins with a molar mass of about 3 to 20 kD (compared to the molar mass of antibodies at ~150 kDa.). Examples of antibody mimetics include affibody molecules affilins, affimers, affitins, alphabodies anticalins, avimers, DARPin,
30 fynomers Kunitz domain peptides and monobodies.

Other Peptides

[0555] In embodiments, the therapeutic moiety includes a peptide. In embodiments, the peptide acts as an agonist, increasing the activity of a target protein. In embodiments, the peptide acts as an antagonist, decreasing the activity of a target protein. In embodiments, the peptide is configured to inhibit protein-protein interaction (PPI). Protein-protein interactions (PPIs) are important in many biochemical processes, including transcription of nucleic acid and various post-translational modifications of translated proteins. PPIs can be experimentally determined by biophysical techniques such as X-ray crystallography, NMR spectroscopy, surface plasma resonance (SPR), bio-layer interferometry (BLI), isothermal titration calorimetry (ITC), radio-ligand binding, spectrophotometric assays and fluorescence spectroscopy. Peptides that inhibit protein-protein interaction can be referred to as peptide inhibitors.

[0556] In embodiments, the therapeutic moiety includes a peptide inhibitor. In embodiments, the peptide inhibitor includes from about 5 to about 100 amino acids, from about 5 to about 50 amino acids; from about 15 to about 30 amino acids; or from about 20 to about 40 amino acids. In embodiments, the peptide inhibitor includes one or more chemical modifications, for example, to reduce proteolytic degradation and/or to improve *in vivo* half-life. In embodiments, the peptide inhibitor includes one or more synthetic amino acids and/or a backbone modification. In embodiments, the peptide inhibitor has an α -helical structure.

[0557] In embodiments, the peptide inhibitor is configured to disrupt one or more function of a protein associated with a disease. In embodiments, the peptide inhibitor is configured to disrupt formation of protein complexes. In embodiments, binding of the peptide inhibitor to the protein blocks dimer formation.

Small Molecules

[0558] In embodiments, the therapeutic moiety includes a small molecule for treating a disease. In embodiments, the small molecule does not readily gain access to an intracellular compartment of a cell when delivered by itself (not conjugated to a delivery construct).

Methods of Making

[0559] The compounds described herein can be prepared in a variety of ways known to one skilled in the art of organic synthesis or variations thereon as appreciated by those skilled in the art. The compounds described herein can be prepared from readily available starting materials. Optimum

reaction conditions can vary with the particular reactants or solvents used, but such conditions can be determined by one skilled in the art. Methods of making the compounds prepared herein are essentially as disclosed in WO 2022/213118, entitled "CYCLIC CELL PENETRATING PEPTIDES", and in WO 2023/205451, entitled "CYCLIC PEPTIDES FOR DELIVERING THERAPEUTICS" the disclosures of which are hereby incorporated by reference in their entireties.

[0560] Variations on the compounds described herein include the addition, subtraction, or movement of the various constituents as described for each compound. Similarly, when one or more chiral centers are present in a molecule, the chirality of the molecule can be changed.

Additionally, compound synthesis can involve the protection and deprotection of various chemical groups. The use of protection and deprotection, and the selection of appropriate protecting groups can be determined by one skilled in the art. The chemistry of protecting groups can be found, for example, in Wuts and Greene, Protective Groups in Organic Synthesis, 4th Ed., Wiley & Sons, 2006, which is incorporated herein by reference in its entirety.

[0561] The starting materials and reagents used in preparing the disclosed compounds and compositions are either available from commercial suppliers such as Aldrich Chemical Co., (Milwaukee, WI), Acros Organics (Morris Plains, NJ), Fisher Scientific (Pittsburgh, PA), Sigma (St. Louis, MO), Pfizer (New York, NY), GlaxoSmithKline (Raleigh, NC), Merck (Whitehouse Station, NJ), Johnson & Johnson (New Brunswick, NJ), Aventis (Bridgewater, NJ), AstraZeneca (Wilmington, DE), Novartis (Basel, Switzerland), Wyeth (Madison, NJ), Bristol-Myers-Squibb (New York, NY), Roche (Basel, Switzerland), Lilly (Indianapolis, IN), Abbott (Abbott Park, IL), Schering Plough (Kenilworth, NJ), or Boehringer Ingelheim (Ingelheim, Germany), or are prepared by methods known to those skilled in the art following procedures set forth in references such as Fieser and Fieser's Reagents for Organic Synthesis, Volumes 1-17 (John Wiley and Sons, 1991); Rodd's Chemistry of Carbon Compounds, Volumes 1-5 and Supplementals (Elsevier Science Publishers, 1989); Organic Reactions, Volumes 1-40 (John Wiley and Sons, 1991); March's Advanced Organic Chemistry, (John Wiley and Sons, 4th Edition); and Larock's Comprehensive Organic Transformations (VCH Publishers Inc., 1989). Other materials, such as the pharmaceutical carriers disclosed herein can be obtained from commercial sources.

[0562] Reactions to produce the compounds described herein can be carried out in solvents, which can be selected by one of skill in the art of organic synthesis. Solvents can be substantially nonreactive with the starting materials (reactants), the intermediates, or products under the conditions at which the reactions are carried out, *i.e.*, temperature and pressure. Reactions can be carried out in one solvent or a mixture of more than one solvent. Product or intermediate formation can be monitored according to any suitable method known in the art. For example, product formation can be monitored by spectroscopic means, such as nuclear magnetic resonance spectroscopy (*e.g.*, ^1H or ^{13}C) infrared spectroscopy, spectrophotometry (*e.g.*, UV-visible), or mass spectrometry, or by chromatography such as high performance liquid chromatography (HPLC) or thin layer chromatography.

[0563] The disclosed compounds can be prepared by solid phase peptide synthesis wherein the amino acid α -N-terminal is protected by an acid or base protecting group. Such protecting groups should have the properties of being stable to the conditions of peptide linkage formation while being readily removable without destruction of the growing peptide chain or racemization of any of the chiral centers contained therein. Suitable protecting groups are 9-fluorenylmethyloxycarbonyl (Fmoc), t-butyloxycarbonyl (Boc), benzyloxycarbonyl (Cbz), biphenylisopropylloxycarbonyl, t-amylloxycarbonyl, isobornyloxycarbonyl, α,α -dimethyl-3,5-dimethoxybenzyloxycarbonyl, o-nitrophenylsulfonyl, 2-cyano-t-butyloxycarbonyl, and the like. The 9-fluorenylmethyloxycarbonyl (Fmoc) protecting group is particularly preferred for the synthesis of the disclosed compounds. Other preferred side chain protecting groups are, for side chain amino groups like lysine and arginine, 2,2,5,7,8-pentamethylchroman-6-sulfonyl (pmc), nitro, p-toluenesulfonyl, 4-methoxybenzene-sulfonyl, Cbz, Boc, and adamantyloxycarbonyl; for tyrosine, benzyl, o-bromobenzyloxy-carbonyl, 2,6-dichlorobenzyl, isopropyl, t-butyl (t-Bu), cyclohexyl, cyclopentyl and acetyl (Ac); for serine, t-butyl, benzyl and tetrahydropyranyl; for histidine, trityl, benzyl, Cbz, p-toluenesulfonyl and 2,4-dinitrophenyl; for tryptophan, formyl, for aspartic acid and glutamic acid, benzyl and t-butyl and for cysteine, triphenylmethyl (trityl). In the solid phase peptide synthesis method, the α -C-terminal amino acid is attached to a suitable solid support or resin. Suitable solid supports useful for the above synthesis are those materials which are inert to the reagents and reaction conditions of the stepwise condensation-deprotection reactions, as well as being insoluble in the media used. Solid supports for synthesis of α -C-terminal carboxy peptides is 4-hydroxymethylphenoxymethyl-copoly(styrene-1%

divinylbenzene) or 4-(2',4'-dimethoxyphenyl-Fmoc-aminomethyl)phenoxyacetamidoethyl resin available from Applied Biosystems (Foster City, Calif.). The α -C-terminal amino acid is coupled to the resin by means of N,N'-dicyclohexylcarbodiimide (DCC), N,N'-diisopropylcarbodiimide (DIC) or O-benzotriazol-1-yl-N,N,N',N'-tetramethyluroniumhexafluorophosphate (HBTU), with or without 4-dimethylaminopyridine (DMAP), 1-hydroxybenzotriazole (HOBT), benzotriazol-1-yl-oxy-tris(dimethylamino)phosphoniumhexafluorophosphate (BOP) or bis(2-oxo-3-oxazolidinyl)phosphine chloride (BOPCl), mediated coupling for from about 1 to about 24 hours at a temperature of between 10°C and 50°C in a solvent such as dichloromethane or DMF. When the solid support is 4-(2',4'-dimethoxyphenyl-Fmoc-aminomethyl)phenoxy-acetamidoethyl resin, the Fmoc group is cleaved with a secondary amine, preferably piperidine, prior to coupling with the α -C-terminal amino acid as described above. One method for coupling to the deprotected 4-(2',4'-dimethoxyphenyl-Fmoc-aminomethyl)phenoxy-acetamidoethyl resin is O-benzotriazol-1-yl-N,N,N',N'-tetramethyluroniumhexafluorophosphate (HBTU, 1 equiv.) and 1-hydroxybenzotriazole (HOBT, 1 equiv.) in DMF. The coupling of successive protected amino acids can be carried out in an automatic polypeptide synthesizer. In one example, the α -N-terminal in the amino acids of the growing peptide chain are protected with Fmoc. The removal of the Fmoc protecting group from the α -N-terminal side of the growing peptide is accomplished by treatment with a secondary amine, preferably piperidine. Each protected amino acid is then introduced in about 3-fold molar excess, and the coupling is preferably carried out in DMF. The coupling agent can be O-benzotriazol-1-yl-N,N,N',N'-tetramethyluroniumhexafluorophosphate (HBTU, 1 equiv.) and 1-hydroxybenzotriazole (HOBT, 1 equiv.). At the end of the solid phase synthesis, the polypeptide is removed from the resin and deprotected, either in successively or in a single operation. Removal of the polypeptide and deprotection can be accomplished in a single operation by treating the resin-bound polypeptide with a cleavage reagent comprising thioanisole, water, ethanedithiol and trifluoroacetic acid. In cases wherein the α -C-terminal of the polypeptide is an alkylamide, the resin is cleaved by aminolysis with an alkylamine. Alternatively, the peptide can be removed by transesterification, *e.g.*, with methanol, followed by aminolysis or by direct transamidation. The protected peptide can be purified at this point or taken to the next step directly. The removal of the side chain protecting groups can be accomplished using the cleavage cocktail described above. The fully deprotected peptide can be purified by a sequence of chromatographic steps employing any or all of the following types: ion

exchange on a weakly basic resin (acetate form); hydrophobic adsorption chromatography on underivatized polystyrene-divinylbenzene (for example, Amberlite XAD); silica gel adsorption chromatography; ion exchange chromatography on carboxymethylcellulose; partition chromatography, *e.g.*, on Sephadex G-25, LH-20 or countercurrent distribution; high
5 performance liquid chromatography (HPLC), especially reverse-phase HPLC on octyl- or octadecylsilyl-silica bonded phase column packing.

[0564] The above polymers, such as PEG groups, can be attached to the cargo under any suitable conditions used to react a protein with an activated polymer molecule. Any means known in the art can be used, including via acylation, reductive alkylation, Michael addition, thiol alkylation
10 or other chemoselective conjugation/ligation methods through a reactive group on the PEG moiety (*e.g.*, an aldehyde, amino, ester, thiol, α -haloacetyl, maleimido or hydrazino group) to a reactive group on the cargo (*e.g.*, an aldehyde, amino, ester, thiol, α -haloacetyl, maleimido or hydrazino group). Activating groups which can be used to link the water soluble polymer to one or more proteins include without limitation sulfone, maleimide, sulfhydryl, thiol, triflate,
15 tresylate, aziridine, oxirane, 5-pyridyl, and alpha-halogenated acyl group (*e.g.*, α -iodo acetic acid, α -bromoacetic acid, α -chloroacetic acid). If attached to the cargo by reductive alkylation, the polymer selected should have a single reactive aldehyde so that the degree of polymerization is controlled. See, for example, Kinstler et al., *Adv. Drug. Delivery Rev.* 54: 477-485 (2002); Roberts et al., *Adv. Drug Delivery Rev.* 54: 459-476 (2002); and Zalipsky et al., *Adv. Drug*
20 *Delivery Rev.* 16: 157-182 (1995).

[0565] In order to directly covalently link the cargo to the CPP, appropriate amino acid residues of the CPP may be reacted with an organic derivatizing agent that is capable of reacting with a selected side chain or the N- or C-termini of an amino acids. Reactive groups on the peptide or conjugate moiety include, *e.g.*, an aldehyde, amino, ester, thiol, α -haloacetyl, maleimido or
25 hydrazino group. Derivatizing agents include, for example, maleimidobenzoyl sulfosuccinimide ester (conjugation through cysteine residues), N-hydroxysuccinimide (through lysine residues), glutaraldehyde, succinic anhydride or other agents known in the art.

[0566] Methods of synthesizing oligomeric antisense compounds are known in the art. The present disclosure is not limited by the method of synthesizing the cargo. In embodiments,
30 provided herein are compounds having reactive phosphorus groups useful for forming internucleoside linkages including for example phosphodiester and phosphorothioate

internucleoside linkages. Methods of preparation and/or purification of precursors or antisense compounds are not a limitation of the compositions or methods provided herein. Methods for synthesis and purification of DNA, RNA, and the antisense compounds are well known to those skilled in the art.

- 5 [0567] Oligomerization of modified and unmodified nucleosides can be routinely performed according to literature procedures for DNA (Protocols for Oligonucleotides and Analogs, Ed. Agrawal (1993), Humana Press) and/or RNA (Scaringe, Methods (2001), 23, 206-217. Gait et al., Applications of Chemically synthesized RNA in RNA: Protein Interactions, Ed. Smith (1998), 1-36. Gallo et al., Tetrahedron (2001), 57, 5707-5713).
- 10 [0568] Antisense compounds provided herein can be conveniently and routinely made through the well-known technique of solid phase synthesis. Equipment for such synthesis is sold by several vendors including, for example, Applied Biosystems (Foster City, CA). Any other means for such synthesis known in the art may additionally or alternatively be employed. It is well known to use similar techniques to prepare oligonucleotides such as the phosphorothioates and
- 15 alkylated derivatives. The invention is not limited by the method of antisense compound synthesis.
- [0569] Methods of oligonucleotide purification and analysis are known to those skilled in the art. Analysis methods include capillary electrophoresis (CE) and electrospray-mass spectroscopy. Such synthesis and analysis methods can be performed in multi-well plates. The method of the
- 20 invention is not limited by the method of oligomer purification.

Methods of Administration

- [0570] *In vivo* application of the disclosed compounds, and compositions containing them, can be accomplished by any suitable method and technique presently or prospectively known to those skilled in the art. For example, the disclosed compounds can be formulated in a
- 25 physiologically- or pharmaceutically-acceptable form and administered by any suitable route known in the art including, for example, oral and parenteral routes of administration. As used herein, the term parenteral includes subcutaneous, intradermal, intravenous, intramuscular, intraperitoneal, intrasternal, intrathecal administration, intracerebroventricular administration, such as by injection. Administration of the disclosed compounds or compositions can be a single
- 30 administration, or at continuous or distinct intervals as can be readily determined by a person skilled in the art.

[0571] The compounds disclosed herein, and compositions comprising them, can also be administered utilizing liposome technology, slow-release capsules, implantable pumps, and biodegradable containers. These delivery methods can, advantageously, provide a uniform dosage over an extended period of time. The compounds can also be administered in their salt derivative forms or crystalline forms.

[0572] The compounds disclosed herein can be formulated according to known methods for preparing pharmaceutically acceptable compositions. Formulations are described in detail in a number of sources which are well known and readily available to those skilled in the art. For example, *Remington's Pharmaceutical Science* by E.W. Martin (1995) describes formulations that can be used in connection with the disclosed methods. In general, the compounds disclosed herein can be formulated such that an effective amount of the compound is combined with a suitable carrier in order to facilitate effective administration of the compound. The compositions used can also be in a variety of forms. These include, for example, solid, semi-solid, and liquid dosage forms, such as tablets, pills, powders, liquid solutions or suspension, suppositories, injectable and infusible solutions, and sprays. The preferred form depends on the intended mode of administration and therapeutic application. The compositions also preferably include conventional pharmaceutically-acceptable carriers and diluents which are known to those skilled in the art. Examples of carriers or diluents for use with the compounds include ethanol, dimethyl sulfoxide, glycerol, alumina, starch, saline, and equivalent carriers and diluents. To provide for the administration of such dosages for the desired therapeutic treatment, compositions disclosed herein can advantageously comprise between about 0.1% and 100% by weight of the total of one or more of the subject compounds based on the weight of the total composition including carrier or diluent.

[0573] Formulations suitable for administration include, for example, aqueous sterile injection solutions, which can contain antioxidants, buffers, bacteriostats, and solutes that render the formulation isotonic with the blood of the intended recipient; and aqueous and nonaqueous sterile suspensions, which can include suspending agents and thickening agents. The formulations can be presented in unit-dose or multi-dose containers, for example sealed ampoules and vials, and can be stored in a freeze dried (lyophilized) condition requiring only the condition of the sterile liquid carrier, for example, water for injections, prior to use. Extemporaneous injection solutions and suspensions can be prepared from sterile powder,

granules, tablets, *etc.* It should be understood that in addition to the ingredients particularly mentioned above, the compositions disclosed herein can include other agents conventional in the art having regard to the type of formulation in question.

[0574] Compounds disclosed herein, and compositions comprising them, can be delivered to a cell either through direct contact with the cell or via a carrier means. Carrier means for delivering compounds and compositions to cells are known in the art and include, for example, encapsulating the composition in a liposome moiety. Another means for delivery of compounds and compositions disclosed herein to a cell comprises attaching the compounds to a protein or nucleic acid that is targeted for delivery to the target cell. U.S. Patent No. 6,960,648 and U.S. Application Publication Nos. 20030032594 and 20020120100 disclose amino acid sequences that can be coupled to another composition and that allows the composition to be translocated across biological membranes. U.S. Application Publication No. 20020035243 also describes compositions for transporting biological moieties across cell membranes for intracellular delivery. Compounds can also be incorporated into polymers, examples of which include poly (D-L lactide-co-glycolide) polymer for intracranial tumors; poly[bis(p-carboxyphenoxy) propane:sebacic acid] in a 20:80 molar ratio (as used in GLIADEL); chondroitin; chitin; and chitosan.

[0575] Compounds and compositions disclosed herein, including pharmaceutically acceptable salts or prodrugs thereof, can be administered intravenously, intramuscularly, or intraperitoneally by infusion or injection. Solutions of the active agent or its salts can be prepared in water, optionally mixed with a nontoxic surfactant. Dispersions can also be prepared in glycerol, liquid polyethylene glycols, triacetin, and mixtures thereof and in oils. Under ordinary conditions of storage and use, these preparations can contain a preservative to prevent the growth of microorganisms.

[0576] The pharmaceutical dosage forms suitable for injection or infusion can include sterile aqueous solutions or dispersions or sterile powders comprising the active ingredient, which are adapted for the extemporaneous preparation of sterile injectable or infusible solutions or dispersions, optionally encapsulated in liposomes. The ultimate dosage form should be sterile, fluid and stable under the conditions of manufacture and storage. The liquid carrier or vehicle can be a solvent or liquid dispersion medium comprising, for example, water, ethanol, a polyol (for example, glycerol, propylene glycol, liquid polyethylene glycols, and the like), vegetable

oils, nontoxic glyceryl esters, and suitable mixtures thereof. The proper fluidity can be maintained, for example, by the formation of liposomes, by the maintenance of the required particle size in the case of dispersions or by the use of surfactants. Optionally, the prevention of the action of microorganisms can be brought about by various other antibacterial and antifungal agents, for example, parabens, chlorobutanol, phenol, sorbic acid, thimerosal, and the like. In many cases, it will be preferable to include isotonic agents, for example, sugars, buffers or sodium chloride. Prolonged absorption of the injectable compositions can be brought about by the inclusion of agents that delay absorption, for example, aluminum monostearate and gelatin.

[0577] Sterile injectable solutions are prepared by incorporating a compound and/or agent disclosed herein in the required amount in the appropriate solvent with various other ingredients enumerated above, as required, followed by filter sterilization. In the case of sterile powders for the preparation of sterile injectable solutions, the preferred methods of preparation are vacuum drying and the freeze drying techniques, which yield a powder of the active ingredient plus any additional desired ingredient present in the previously sterile-filtered solutions.

[0578] Useful dosages of the compounds and agents and pharmaceutical compositions disclosed herein can be determined by comparing their *in vitro* activity, and *in vivo* activity in animal models. Methods for the extrapolation of effective dosages in mice, and other animals, to humans are known to the art.

[0579] The dosage ranges for the administration of the compositions are those large enough to produce the desired effect in which the symptoms or disorder are affected. The dosage should not be so large as to cause adverse side effects, such as unwanted cross-reactions, anaphylactic reactions, and the like. Generally, the dosage will vary with the age, condition, sex and extent of the disease in the patient and can be determined by one of skill in the art. The dosage can be adjusted by the individual physician in the event of any counterindications. Dosage can vary, and can be administered in one or more dose administrations daily, for one or several days.

[0580] Also disclosed are pharmaceutical compositions that comprise a compound disclosed herein in combination with a pharmaceutically acceptable carrier. Pharmaceutical compositions adapted for oral, topical or parenteral administration, comprising an amount of a compound constitute a preferred aspect. The dose administered to a patient, particularly a human, should be sufficient to achieve a therapeutic response in the patient over a reasonable time frame, without lethal toxicity, and preferably causing no more than an acceptable level of side effects or

morbidity. One skilled in the art will recognize that dosage will depend upon a variety of factors including the condition (health) of the subject, the body weight of the subject, kind of concurrent treatment, if any, frequency of treatment, therapeutic ratio, as well as the severity and stage of the pathological condition.

5 [0581] Also disclosed are kits that comprise a compound disclosed herein in one or more containers. The disclosed kits can optionally include pharmaceutically acceptable carriers and/or diluents. In one embodiment, a kit includes one or more other components, adjuncts, or adjuvants as described herein. In another embodiment, a kit includes one or more anti-cancer agents, such as those agents described herein. In one embodiment, a kit includes instructions or packaging
10 materials that describe how to administer a compound or composition of the kit. Containers of the kit can be of any suitable material, *e.g.*, glass, plastic, metal, *etc.*, and of any suitable size, shape, or configuration. In one embodiment, a compound and/or agent disclosed herein is provided in the kit as a solid, such as a tablet, pill, or powder form. In another embodiment, a compound and/or agent disclosed herein is provided in the kit as a liquid or solution. In one
15 embodiment, the kit comprises an ampoule or syringe containing a compound and/or agent disclosed herein in liquid or solution form.

[0582] A number of embodiments of the invention have been described. Nevertheless, it will be understood that various modifications may be made without departing from the spirit and scope of the invention. Accordingly, other embodiments are within the scope of the following claims.

20 **Certain Definitions**

[0583] As used in the description and the appended claims, the singular forms “a” “an” and “the” include plural referents unless the context clearly dictates otherwise. Thus, for example, reference to “a composition” includes mixtures of two or more such compositions, reference to “an agent” includes mixtures of two or more such agents, reference to “the component” includes
25 mixtures of two or more such components, and the like.

[0584] The term “about” when immediately preceding a numerical value means a range (*e.g.*, plus or minus 10% of that value). For example, “about 50” can mean 45 to 55, “about 25,000” can mean 22,500 to 27,500, *etc.*, unless the context of the disclosure indicates otherwise, or is inconsistent with such an interpretation. For example, in a list of numerical values such as “about
30 49, about 50, about 55, ...”, “about 50” means a range extending to less than half the interval(s) between the preceding and subsequent values, *e.g.*, more than 49.5 to less than 52.5.

Furthermore, the phrases “less than about” a value or “greater than about” a value should be understood in view of the definition of the term “about” provided herein. Similarly, the term “about” when preceding a series of numerical values or a range of values (e.g., “about 10, 20, 30” or “about 10-30”) refers, respectively, to all values in the series, or the endpoints of the range.

[0585] As used herein, “polyethylene glycol” and “PEG” are used interchangeably. PEG refers to the repeat group of the chemical formula $-\text{CH}_2\text{CH}_2\text{O}-$.

[0586] As used herein, the term “cell penetrating peptide”, “CPP”, “cyclic cell penetrating peptide” or “cCPP” refers to a peptide that facilitates the delivery of a cargo to the cytosol of a cell.

[0587] As used herein, the term “endosomal escape vehicle” (EEV) refers to a cCPP that is conjugated by a chemical linkage (i.e., a covalent bond or non-covalent interaction) to a linker and/or an exocyclic peptide (EP).

[0588] As used herein, the term “EEV-cargo conjugate” refers to an endosomal escape vehicle defined herein conjugated by a chemical linkage (i.e., a covalent bond or non-covalent interaction) to a cargo. The cargo can be delivered into a cell by the EEV.

[0589] As used herein, the term “exocyclic peptide” (EP) refers to two or more amino acid residues linked by a peptide bond that can be conjugated to a cyclic cell penetrating peptide (cCPP) disclosed herein. The EP, when conjugated to a cyclic peptide, may alter the tissue distribution and/or retention of the compound. Typically, the EP comprises at least one positively charged amino acid residue, e.g., at least one lysine residue and/or at least one arginine residue. Non-limiting examples of EP are described herein. The EP can be a peptide that has been identified in the art as a “nuclear localization sequence” (NLS).

[0590] As used herein, “linker” or “L” refers to a moiety that covalently bonds one or more moieties (e.g., an exocyclic peptide (EP) and an AC to the cyclic cell penetrating peptide (cCPP)). The linker can comprise a natural or non-natural amino acid or polypeptide. The linker can be a synthetic compound containing two or more appropriate functional groups suitable to bind the cCPP to an AC, to thereby form the compounds disclosed herein. The linker can comprise a polyethylene glycol (PEG) moiety. The linker can comprise one or more amino acids. The cCPP may be covalently bound to a cargo and/or an exocyclic peptide (EP) via a linker.

[0591] As used herein, the term “oligonucleotide” refers to an oligomeric compound comprising a plurality of linked nucleotides or nucleosides. One or more nucleotides of an oligonucleotide can be modified. An oligonucleotide can comprise ribonucleic acid (RNA) or deoxyribonucleic acid (DNA). Oligonucleotides can be composed of natural and/or modified nucleobases, sugars and covalent internucleoside linkages, and can further include non-nucleic acid conjugates.

[0592] The terms “peptide,” “protein,” and “polypeptide” are used interchangeably to refer to a natural or synthetic molecule comprising two or more amino acids linked by the carboxyl group of one amino acid to the alpha amino group of another. Two or more amino acid residues can be linked by the carboxyl group of one amino acid to the alpha amino group. Two or more amino acids of the polypeptide can be joined by a peptide bond. The polypeptide can include a peptide backbone modification in which two or more amino acids are covalently attached by a bond other than a peptide bond. The polypeptide can include one or more non-natural amino acids, amino acid analogs, or other synthetic molecules that are capable of integrating into a polypeptide. The term polypeptide includes naturally occurring and artificially occurring amino acids. The term polypeptide includes peptides, for example, that include from about 2 to about 100 amino acid residues as well as proteins, that include more than about 100 amino acid residues, or more than about 1000 amino acid residues, including, but not limited to therapeutic proteins such as antibodies, enzymes, receptors, soluble proteins and the like.

[0593] The term “therapeutic polypeptide” refers to a polypeptide that has therapeutic, prophylactic or other biological activity. The therapeutic polypeptide can be produced in any suitable manner. For example, the therapeutic polypeptide may be isolated or purified from a naturally occurring environment, may be chemically synthesized, may be recombinantly produced, or a combination thereof.

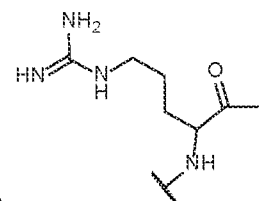
[0594] The term “small molecule” refers to an organic compound with pharmacological activity and a molecular weight of less than about 2000 Daltons, or less than about 1000 Daltons, or less than about 500 Daltons. Small molecule therapeutics are typically manufactured by chemical synthesis.

[0595] As used herein, the term “contiguous” refers to two amino acids, which are connected by a covalent bond. For example, in the context of a representative cyclic cell penetrating peptide



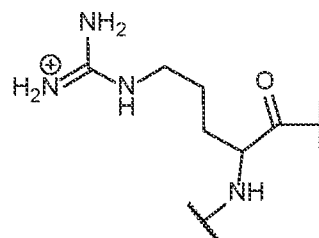
(cCPP) such as , AA₁/AA₂, AA₂/AA₃, AA₃/AA₄, and AA₅/AA₁ exemplify pairs of contiguous amino acids.

[0596] A residue of a chemical species, as used herein, refers to a derivative of the chemical species that is present in a particular product. To form the product, at least one atom of the species is replaced by a bond to another moiety, such that the product contains a derivative, or residue, of the chemical species. For example, the cyclic cell penetrating peptides (cCPP) described herein have amino acids (e.g., arginine) incorporated therein through formation of one or more peptide bonds. The amino acids incorporated into the cCPP may be referred to residues,



or simply as an amino acid. Thus, arginine or an arginine residue refers to

[0597] The term “protonated form thereof” refers to a protonated form of an amino acid. For example, the guanidine group on the side chain of arginine may be protonated to form a



guanidinium group. The structure of a protonated form of arginine is

[0598] As used herein, the term “chirality” refers to a molecule that has more than one stereoisomer that differs in the three-dimensional spatial arrangement of atoms, in which one stereoisomer is a non-superimposable mirror image of the other. Amino acids, except for glycine, have a chiral carbon atom adjacent to the carboxyl group. The term “enantiomer” refers to stereoisomers that are chiral. The chiral molecule can be an amino acid residue having a “D” and “L” enantiomer. Molecules without a chiral center, such as glycine, can be referred to as “achiral.”

[0599] As used herein, the term “hydrophobic” refers to a moiety that is not soluble in water or has minimal solubility in water. Generally, neutral moieties and/or non-polar moieties, or

moieties that are predominately neutral and/or non-polar are hydrophobic. Hydrophobicity can be measured by one of the methods disclosed herein below.

[0600] As used herein “aromatic” refers to an unsaturated cyclic molecule having $4n + 2 \pi$ electrons, wherein n is any integer. The term “non-aromatic” refers to any unsaturated cyclic molecule which does not fall within the definition of aromatic.

[0601] “Alkyl”, “alkyl chain” or “alkyl group” refer to a fully saturated, straight, or branched hydrocarbon chain radical having from one to forty carbon atoms, and which is attached to the rest of the molecule by a single bond. Alkyls comprising any number of carbon atoms from 1 to 40 are included. An alkyl comprising up to 40 carbon atoms is a C_1 - C_{40} alkyl, an alkyl comprising up to 10 carbon atoms is a C_1 - C_{10} alkyl, an alkyl comprising up to 6 carbon atoms is a C_1 - C_6 alkyl and an alkyl comprising up to 5 carbon atoms is a C_1 - C_5 alkyl. A C_1 - C_5 alkyl includes C_5 alkyls, C_4 alkyls, C_3 alkyls, C_2 alkyls and C_1 alkyl (*i.e.*, methyl). A C_1 - C_6 alkyl includes all moieties described above for C_1 - C_5 alkyls but also includes C_6 alkyls. A C_1 - C_{10} alkyl includes all moieties described above for C_1 - C_5 alkyls and C_1 - C_6 alkyls, but also includes C_7 , C_8 , C_9 and C_{10} alkyls. Similarly, a C_1 - C_{12} alkyl includes all the foregoing moieties, but also includes C_{11} and C_{12} alkyls. Non-limiting examples of C_1 - C_{12} alkyl include methyl, ethyl, *n*-propyl, *i*-propyl, *sec*-propyl, *n*-butyl, *i*-butyl, *sec*-butyl, *t*-butyl, *n*-pentyl, *t*-amyl, *n*-hexyl, *n*-heptyl, *n*-octyl, *n*-nonyl, *n*-decyl, *n*-undecyl, and *n*-dodecyl. Unless stated otherwise specifically in the specification, an alkyl group can be optionally substituted.

[0602] “Alkylene”, “alkylene chain” or “alkylene group” refers to a fully saturated, straight or branched divalent hydrocarbon chain radical, having from one to forty carbon atoms. Non-limiting examples of C_2 - C_{40} alkylene include ethylene, propylene, *n*-butylene, ethenylene, propenylene, *n*-butenylene, propynylene, *n*-butynylene, and the like. Unless stated otherwise specifically in the specification, an alkylene chain can be optionally substituted.

[0603] “Alkenyl”, “alkenyl chain” or “alkenyl group” refers to a straight or branched hydrocarbon chain radical having from two to forty carbon atoms and having one or more carbon-carbon double bonds. Each alkenyl group is attached to the rest of the molecule by a single bond. Alkenyl groups comprising any number of carbon atoms from 2 to 40 are included. An alkenyl group comprising up to 40 carbon atoms is a C_2 - C_{40} alkenyl, an alkenyl comprising up to 10 carbon atoms is a C_2 - C_{10} alkenyl, an alkenyl group comprising up to 6 carbon atoms is a C_2 - C_6 alkenyl and an alkenyl comprising up to 5 carbon atoms is a C_2 - C_5 alkenyl. A C_2 - C_5

alkenyl includes C₅ alkenyls, C₄ alkenyls, C₃ alkenyls, and C₂ alkenyls. A C₂-C₆ alkenyl includes all moieties described above for C₂-C₅ alkenyls but also includes C₆ alkenyls. A C₂-C₁₀ alkenyl includes all moieties described above for C₂-C₅ alkenyls and C₂-C₆ alkenyls, but also includes C₇, C₈, C₉ and C₁₀ alkenyls. Similarly, a C₂-C₁₂ alkenyl includes all the foregoing moieties, but also includes C₁₁ and C₁₂ alkenyls. Non-limiting examples of C₂-C₁₂ alkenyl include ethenyl (vinyl), 1-propenyl, 2-propenyl (allyl), iso-propenyl, 2-methyl-1-propenyl, 1-butenyl, 2-butenyl, 3-butenyl, 1-pentenyl, 2-pentenyl, 3-pentenyl, 4-pentenyl, 1-hexenyl, 2-hexenyl, 3-hexenyl, 4-hexenyl, 5-hexenyl, 1-heptenyl, 2-heptenyl, 3-heptenyl, 4-heptenyl, 5-heptenyl, 6-heptenyl, 1-octenyl, 2-octenyl, 3-octenyl, 4-octenyl, 5-octenyl, 6-octenyl, 7-octenyl, 1-nonenyl, 2-nonenyl, 3-nonenyl, 4-nonenyl, 5-nonenyl, 6-nonenyl, 7-nonenyl, 8-nonenyl, 1-decenyl, 2-decenyl, 3-decenyl, 4-decenyl, 5-decenyl, 6-decenyl, 7-decenyl, 8-decenyl, 9-decenyl, 1-undecenyl, 2-undecenyl, 3-undecenyl, 4-undecenyl, 5-undecenyl, 6-undecenyl, 7-undecenyl, 8-undecenyl, 9-undecenyl, 10-undecenyl, 1-dodecenyl, 2-dodecenyl, 3-dodecenyl, 4-dodecenyl, 5-dodecenyl, 6-dodecenyl, 7-dodecenyl, 8-dodecenyl, 9-dodecenyl, 10-dodecenyl, and 11-dodecenyl. Unless stated otherwise specifically in the specification, an alkyl group can be optionally substituted.

[0604] “Alkenylene”, “alkenylene chain” or “alkenylene group” refers to a straight or branched divalent hydrocarbon chain radical, having from two to forty carbon atoms, and having one or more carbon-carbon double bonds. Non-limiting examples of C₂-C₄₀ alkenylene include ethene, propene, butene, and the like. Unless stated otherwise specifically in the specification, an alkenylene chain can be optionally.

[0605] “Acetyl” (Ac-) refers to the group CH₃C(O)-.

[0606] “Alkoxy” or “alkoxy group” refers to the group -OR, where R is alkyl, alkenyl, alkynyl, cycloalkyl, or heterocyclyl as defined herein. Unless stated otherwise specifically in the specification, an alkoxy group can be optionally substituted.

[0607] “Acyl” or “acyl group” refers to groups -C(O)R, where R is hydrogen, alkyl, alkenyl, alkynyl, carbocyclyl, or heterocyclyl, as defined herein. Unless stated otherwise specifically in the specification, acyl can be optionally substituted.

[0608] “Alkylcarbamoyl” or “alkylcarbamoyl group” refers to the group -O-C(O)-NR_aR_b, where R_a and R_b are the same or different and are independently an alkyl, alkenyl, alkynyl, aryl, heteroaryl, as defined herein, or R_aR_b can be taken together to form a cycloalkyl group or

heterocyclyl group, as defined herein. Unless stated otherwise specifically in the specification, an alkylcarbamoyl group can be optionally substituted.

[0609] “Alkylcarboxamidyl” or “alkylcarboxamidyl group” refers to the group $-C(O)-NR_aR_b$, where R_a and R_b are the same or different and are independently an alkyl, alkenyl, alkynyl, aryl, heteroaryl, cycloalkyl, cycloalkenyl, cycloalkynyl, or heterocyclyl group, as defined herein, or R_aR_b can be taken together to form a cycloalkyl group, as defined herein. Unless stated otherwise specifically in the specification, an alkylcarboxamidyl group can be optionally substituted.

[0610] “Aryl” refers to a hydrocarbon ring system radical comprising hydrogen, 6 to 18 carbon atoms and at least one aromatic ring. For purposes of this invention, the aryl radical can be a monocyclic, bicyclic, tricyclic or tetracyclic ring system, which can include fused or bridged ring systems. Aryl radicals include, but are not limited to, aryl radicals derived from aceanthrylene, acenaphthylene, acephenanthrylene, anthracene, azulene, benzene, chrysene, fluoranthene, fluorene, *as*-indacene, *s*-indacene, indane, indene, naphthalene, phenalene, phenanthrene, pleiadene, pyrene, and triphenylene. Unless stated otherwise specifically in the specification, the term “aryl” is meant to include aryl radicals that are optionally substituted.

[0611] “Heteroaryl” refers to a 5- to 20-membered ring system radical comprising hydrogen atoms, one to thirteen carbon atoms, one to six heteroatoms selected from nitrogen, oxygen and sulfur, and at least one aromatic ring. For purposes of this invention, the heteroaryl radical can be a monocyclic, bicyclic, tricyclic or tetracyclic ring system, which can include fused or bridged ring systems; and the nitrogen, carbon or sulfur atoms in the heteroaryl radical can be optionally oxidized; the nitrogen atom can be optionally quaternized. Examples include, but are not limited to, azepinyl, acridinyl, benzimidazolyl, benzothiazolyl, benzindolyl, benzodioxolyl, benzofuranyl, benzooxazolyl, benzothiazolyl, benzothiadiazolyl, benzo[*b*][1,4]dioxepinyl, 1,4-benzodioxanyl, benzonaphthofuranyl, benzoxazolyl, benzodioxolyl, benzodioxinyl, benzopyranyl, benzopyranonyl, benzofuranyl, benzofuranonyl, benzothienyl (benzothiophenyl), benzotriazolyl, benzo[4,6]imidazo[1,2-*a*]pyridinyl, carbazolyl, cinnoliny, dibenzofuranyl, dibenzothiophenyl, furanyl, furanonyl, isothiazolyl, imidazolyl, indazolyl, indolyl, indazolyl, isoindolyl, indoliny, isoindoliny, isoquinolyl, indoliziny, isoxazolyl, naphthyridinyl, oxadiazolyl, 2-oxoazepinyl, oxazolyl, oxiranyl, 1-oxidopyridinyl, 1-oxidopyrimidinyl, 1-oxidopyrazinyl, 1-oxidopyridazinyl, 1-phenyl-1*H*-pyrrolyl, phenazinyl, phenothiazinyl, phenoxazinyl, phthalazinyl, pteridinyl, purinyl, pyrrolyl, pyrazolyl, pyridinyl, pyrazinyl,

pyrimidinyl, pyridazinyl, quinazolinyl, quinoxalinyl, quinolinyl, quinuclidinyl, isoquinolinyl, tetrahydroquinolinyl, thiazolyl, thiadiazolyl, triazolyl, tetrazolyl, triazinyl, and thiophenyl (i.e., thienyl). Unless stated otherwise specifically in the specification, a heteroaryl group can be optionally substituted.

5 [0612] “Bip” is 3-(4',4'-biphenyl)-L-alanine.

[0613] As used herein, “polyethylene glycol” and “PEG” are used interchangeably. (PEG)_x' or (PEG)_z' refer to a repeat group of the chemical formula -CH₂CH₂O-.

[0614] The term “substituted” used herein means any of the above groups (i.e., alkyl, alkenyl, alkynyl, cycloalkyl, cycloalkenyl, cycloalkynyl, heterocyclyl, aryl, heteroaryl, alkoxy, aryloxy, acyl, alkylcarbamoyl, alkylcarboxamidyl, alkoxycarbonyl, alkylthio, or arylthio) wherein at least
 10 one atom is replaced by a non-hydrogen atoms such as, but not limited to: a halogen atom such as F, Cl, Br, and I; an oxygen atom in groups such as hydroxyl groups, alkoxy groups, and ester groups; a sulfur atom in groups such as thiol groups, thioalkyl groups, sulfone groups, sulfonyl groups, and sulfoxide groups; a nitrogen atom in groups such as amines, amides, alkylamines, dialkylamines, arylamines, alkylarylamines, diarylamines, N-oxides, imides, and enamines; a
 15 silicon atom in groups such as trialkylsilyl groups, dialkylarylsilyl groups, alkylidiarylsilyl groups, and triarylsilyl groups; and other heteroatoms in various other groups. “Substituted” also means any of the above groups in which one or more atoms are replaced by a higher-order bond (e.g., a double- or triple-bond) to a heteroatom such as oxygen in oxo, carbonyl, carboxyl, and
 20 ester groups; and nitrogen in groups such as imines, oximes, hydrazones, and nitriles. For example, “substituted” includes any of the above groups in which one or more atoms are replaced with -NR_gR_h, -NR_gC(=O)R_h, -NR_gC(=O)NR_gR_h, -NR_gC(=O)OR_h, -NR_gSO₂R_h, -OC(=O)NR_gR_h, -OR_g, -SR_g, -SOR_g, -SO₂R_g, -OSO₂R_g, -SO₂OR_g, =NSO₂R_g, and -SO₂NR_gR_h.
 “Substituted also means any of the above groups in which one or more hydrogen atoms are
 25 replaced with -C(=O)R_g, -C(=O)OR_g, -C(=O)NR_gR_h, -CH₂SO₂R_g, -CH₂SO₂NR_gR_h. In the foregoing, R_g and R_h are the same or different and independently hydrogen, alkyl, alkenyl, alkynyl, alkoxy, alkylamino, thioalkyl, aryl, aralkyl, cycloalkyl, cycloalkenyl, cycloalkynyl, cycloalkylalkyl, haloalkyl, haloalkenyl, haloalkynyl, heterocyclyl, *N*-heterocyclyl, heterocyclylalkyl, heteroaryl, *N*-heteroaryl and/or heteroarylalkyl. “Substituted” further means
 30 any of the above groups in which one or more atoms are replaced by an amino, cyano, hydroxyl, imino, nitro, oxo, thioxo, halo, alkyl, alkenyl, alkynyl, alkoxy, alkylamino, thioalkyl, aryl,

aralkyl, cycloalkyl, cycloalkenyl, cycloalkynyl, cycloalkylalkyl, haloalkyl, haloalkenyl, haloalkynyl, heterocyclyl, *N*-heterocyclyl, heterocyclylalkyl, heteroaryl, *N*-heteroaryl and/or heteroarylalkyl group. "Substituted" can also mean an amino acid in which one or more atoms on the side chain are replaced by alkyl, alkenyl, alkynyl, acyl, alkylcarboxamidyl, alkoxy carbonyl, carbocyclyl, heterocyclyl, aryl, or heteroaryl. In addition, each of the foregoing substituents can also be optionally substituted with one or more of the above substituents.

[0615] As used herein, by a "subject" is meant an individual. Thus, the "subject" can include domesticated animals (*e.g.*, cats, dogs, etc.), livestock (*e.g.*, cattle, horses, pigs, sheep, goats, etc.), laboratory animals (*e.g.*, mouse, rabbit, rat, guinea pig, etc.), and birds. "Subject" can also include a mammal, such as a primate or a human. Thus, the subject can be a human or veterinary patient. The term "patient" refers to a subject under the treatment of a clinician, *e.g.*, physician.

[0616] The terms "inhibit", "inhibiting" or "inhibition" refer to a decrease in an activity, expression, function or other biological parameter and can include, but does not require complete ablation of the activity, expression, function or other biological parameter. Inhibition can include, for example, at least about a 10% reduction in the activity, response, condition, or disease as compared to a control. In embodiments, expression, activity or function of a gene or protein is decreased by a statistically significant amount. Thus, the reduction can be a 10, 20, 30, 40, 50, 60, 70, 80, 90, 100%, or any amount of reduction in between as compared to native or control levels.

[0617] By "reduce" or other forms of the word, such as "reducing" or "reduction," is meant lowering of an event or characteristic (*e.g.*, tumor growth). It is understood that this is typically in relation to some standard or expected value, in other words it is relative, but that it is not always necessary for the standard or relative value to be referred to. For example, "reduces tumor growth" means reducing the rate of growth of a tumor relative to a standard or a control (*e.g.*, an untreated tumor).

[0618] The term "treatment" refers to the medical management of a patient with the intent to cure, ameliorate, stabilize, or prevent a disease, pathological condition, or disorder. This term includes active treatment, that is, treatment directed specifically toward the improvement of a disease, pathological condition, or disorder, and also includes causal treatment, that is, treatment directed toward removal of the cause of the associated disease, pathological condition, or disorder. In addition, this term includes palliative treatment, that is, treatment designed for the

relief of symptoms rather than the curing of the disease, pathological condition, or disorder; preventative treatment, that is, treatment directed to minimizing or partially or completely inhibiting the development of the associated disease, pathological condition, or disorder; and supportive treatment, that is, treatment employed to supplement another specific therapy directed toward the improvement of the associated disease, pathological condition, or disorder.

[0619] The term “therapeutically effective” refers to the amount of the composition used is of sufficient quantity to ameliorate one or more causes or symptoms of a disease or disorder. Such amelioration only requires a reduction or alteration, not necessarily elimination.

[0620] The term “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 problems or complications commensurate with a reasonable benefit/risk ratio.

[0621] The term “carrier” means a compound, composition, substance, or structure that, when in combination with a compound or composition, aids or facilitates preparation, storage, administration, delivery, effectiveness, selectivity, or any other feature of the compound or composition for its intended use or purpose. For example, a carrier can be selected to minimize any degradation of the active ingredient and to minimize any adverse side effects in the subject.

[0622] As used herein, the term “pharmaceutically acceptable carrier” refers to sterile aqueous or nonaqueous solutions, dispersions, suspensions or emulsions, as well as sterile powders for reconstitution into sterile injectable solutions or dispersions just prior to use. Examples of suitable aqueous and nonaqueous carriers, diluents, solvents or vehicles include water, ethanol, polyols (such as glycerol, propylene glycol, polyethylene glycol and the like), carboxymethylcellulose and suitable mixtures thereof, vegetable oils (such as olive oil) and injectable organic esters such as ethyl oleate. Proper fluidity can be maintained, for example, by the use of coating materials such as lecithin, by the maintenance of the required particle size in the case of dispersions and by the use of surfactants. These compositions can also contain adjuvants such as preservatives, wetting agents, emulsifying agents and dispersing agents.

Prevention of the action of microorganisms can be ensured by the inclusion of various antibacterial and antifungal agents such as paraben, chlorobutanol, phenol, sorbic acid and the like. It can also be desirable to include isotonic agents such as sugars, sodium chloride and the

like. The injectable formulations can be sterilized, for example, by filtration through a bacterial-retaining filter or by incorporating sterilizing agents in the form of sterile solid compositions which can be dissolved or dispersed in sterile water or other sterile injectable media just prior to use. Suitable inert carriers can include sugars such as lactose.

5 [0623] As used herein, the terms “antisense compound” and “AC” are used interchangeably to refer to a polymeric nucleic acid structure (which can also be referred to as an oligonucleotide or polynucleotide) which is at least partially complementary to a target nucleic acid molecule to which it (the AC) hybridizes. The AC may be a short (in embodiments, less than 50 base pair) polynucleotide or polynucleotide homologue comprising a sequence complimentary to a target
10 sequence in a target pre-mRNA strand. The AC may be formed of natural nucleic acids, synthetic nucleic acids, nucleic acid homologues, or any combination thereof. In embodiments, the AC comprises oligonucleosides. In embodiments, AC comprises antisense oligonucleotides. In embodiments, the AC comprises conjugate groups. Nonlimiting examples of ACs include, but are not limited to, primers, probes, antisense oligonucleotides, external guide sequence (EGS)
15 oligonucleotides, alternate splicers, siRNAs, oligonucleotides, oligonucleosides, oligonucleotide analogs, oligonucleotide mimetics, and chimeric combinations of these. As such, these compounds can be introduced in the form of single-stranded, double-stranded, circular, branched or hairpins and can contain structural elements such as internal or terminal bulges or loops. Oligomeric double-stranded compounds can be two strands hybridized to form double-stranded
20 compounds or a single strand with sufficient self-complementarity to allow for hybridization and formation of a fully or partially double-stranded compound. In embodiments, an AC modulates (increases, decreases, or changes) expression of a target nucleic acid. Various modifications may be made to the polymeric nucleic acid structure, such as phosphorodiamidate morpholino (PMO). Therefore, AC as used herein encompasses any modification described herein, such as a
25 PMO.

[0624] As used herein, the term “oligonucleotide” refers to an oligomeric compound comprising a plurality of linked nucleotides or nucleosides. In certain embodiment, one or more nucleotides of an oligonucleotide is modified. In embodiments, an oligonucleotide comprises ribonucleic acid (RNA) or deoxyribonucleic acid (DNA). In embodiments, oligonucleotides are composed of
30 natural and/or modified nucleobases, sugars and covalent internucleoside linkages, and may further include non-nucleic acid conjugates.

[0625]

[0626] A number of embodiments of the invention have been described. Nevertheless, it will be understood that various modifications may be made without departing from the spirit and scope of the invention. Other embodiments are within the scope of the following claims.

- 5 [0627] All publications, patents and patent applications mentioned in the specification are indicative of the level of skill of those skilled in the art to which this invention pertains. All publications, patents and patent applications are herein incorporated by reference to the same extent as if each individual publication or patent application was specifically and individually indicated to be incorporated by reference.

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EXAMPLES

[0628] While EEV sequences are listed in some Tables, it is understood that the EEV sequences can be conjugated to PMO using amide or click chemistry.

Example 1. Effects on Efficacy and Tolerability of Linker and Conjugation Chemistry

- 15 [0629] This study employs a mouse model (MDX/D2-C) to study the effect on delivery efficacy of a linker that has been installed using strain-promoted alkyne azide cycloaddition (SPAAC) compared to the effect on delivery efficacy of a linker that has been installed by amide conjugation.

[0630] Two compounds were synthesized having the general formula:

Ac-PKKKRKV-PEG₂-K(*cyclo*[Ff-Nal-Cit-r-Cit-r-Q])-Linker-PMO

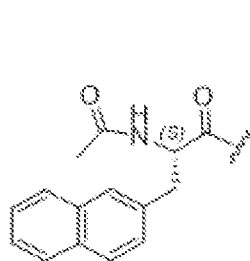
- 20 wherein “EEV1-PMO23-A” and “EEV1-PMO23-B” each comprise a different linker. The linker for EEV1-PMO23-A is installed using amide conjugation to give the amide shown in FIG. 1, whereas the linker for EEV1-PMO23-B is installed using SPAAC to give the trizazinyl group shown in FIG. 1. The sequence for PMO23 is also shown in FIG. 1.

- 25 [0631] EEV1-PMO23-A, EEV1-PMO23-B, and PMO23 (unconjugated and unfunctionalized PMO) were dissolved in saline and injected into mdx mice at a dose of 30 mg/kg PMO equivalent at a volume of 5 mL/kg. Seven days post-injection, the animals were sacrificed, the indicated tissues (i.e., triceps, TiA, heart, and diaphragm) were harvested, snap-frozen, and pulverized. After mRNA extraction, each tissue sample was analyzed for exon 23 skipping by reverse transcription-polymerase chain reaction (RT-PCR) and quantified by densitometry. This
30 experiment demonstrated that EEV1-PMO23-B, which has a linker installed via SPAAC, has

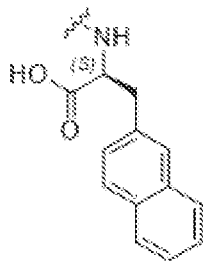
enhanced potency across tissues relative to the EEV1-PMO23-A conjugate having a linker installed via amide conjugation. While not wishing to be bound by theory, it is believed that the reason for the higher potency of EEV1-PMO23-B is that the SPAAC-installed linker includes a hydrophobic component, whereas the amide-conjugation-installed linker does not, resulting in higher overall hydrophobicity for the conjugate EEV1-PMO23-B.

Example 2. Including hydrophobic component in linker

[0632] EEV2-PMO23, EEV3-PMO23, EEV4-PMO23, EEV5-PMO23, and EEV6-PMO23, the structures of each of which is shown in in FIG. 2, were tested using essentially the same procedure as described in Example 1. The structures for EEV3-PMO23, EEV4-PMO23, and EEV5-PMO23 are represented in cartoon form in FIG. 3. Each of these compounds was synthesized via amide conjugation. Each compound was dissolved in saline and injected into mdx mice at a dose of 40 mg/kg at a volume of 5 mL/kg. Seven days post-injection, the animals were sacrificed, the indicated tissues were harvested, snap-frozen, and pulverized. Following protein extraction, dystrophin protein expression was quantified by capillary electrophoresis and represented relative to dystrophin levels in the tissue of a wild-type mouse. This experiment demonstrated that incorporating linkers with and without hydrophobic C-terminal residues of the formula:



in EEV4 and



in EEV 5,

enhanced efficacy as determined by restoration of dystrophin expression across skeletal and cardiac muscle. EEV3 provides an alternative linker for cyclic peptide conjugation that confers increased proteolytic stability and moderate hydrophobicity. EEV4 comprises a Lys-sidechain functionalized hydrophobic component. EEV5 comprises a linker with a hydrophobic residue in the main chain.

Example 3. C-Terminal Lys-side chain hydrophobic derivatives

[0633] EEV2-PMO23, EEV4-PMO23, EEV7- PMO23, EEV8-PMO23, EEV9-PMO23, EEV10-PMO23, EEV11-PMO23, EEV12-PMO23, and EEV13-PMO23, the structures of each of which is shown in in FIG. 4, were tested using essentially the same procedure as described in Example 1. Each of these compounds was synthesized via amide conjugation. Each compound was dissolved in saline and injected into mdx mice at a dose of 40 mg/kg at a volume of 5 mL/kg. Seven days post-injection, the animals were sacrificed, the indicated tissues were harvested, snap-frozen, and pulverized. Following protein extraction, dystrophin protein expression was quantified by capillary electrophoresis and represented relative to dystrophin levels in the tissue of a wild-type mouse. This experiment demonstrated that compounds comprising in which the linker included a C-terminal Lys with a hydrophobic components appended to the side chain had enhanced efficacy as determined by restoration of dystrophin expression in skeletal and cardiac muscle.

Example 4. C-Terminal main-chain hydrophobic derivatives

[0634] EEV2-PMO23, EEV5-PMO23, EEV14- PMO23, EEV15-PMO23, EEV16-PMO23, EEV17-PMO23, EEV18-PMO23, EEV19-PMO23, EEV20-PMO23, and EEV25-PMO23, the structures of each of which is shown in in FIG. 5, were tested using essentially the same procedure as described in Example 1. Each of these compounds was synthesized via amide conjugation. Each compound was dissolved in saline and injected into mdx mice at a dose of 40 mg/kg at a volume of 5 mL/kg. Seven days post-injection, the animals were sacrificed, the indicated tissues were harvested, snap-frozen, and pulverized. Following protein extraction, dystrophin protein expression was quantified by capillary electrophoresis and represented relative to dystrophin levels in the tissue of a wild-type mouse. This experiment demonstrated that incorporating hydrophobic amino acids in the linker backbone enhanced efficacy as determined by restoration of dystrophin expression in skeletal and cardiac muscle.

Example 5. Design and synthesis of EEV

[0635] A phosphorodiamidate morpholino oligomer (PMO) was designed to target a missense EGFP gene with a mutation introduced at nucleotide 654 of intron 2 of the human β globin gene. The mutation at nucleotide 654 activates aberrant splice sites and results in retention of the intron fragment in spliced, mature mRNA, preventing proper translation of EGFP. The EGFP-PMO (5'-

GCTATTACCTTAACCCAG-3') was designed to bind to and block the aberrant splice site in order to correct pre-mRNA splicing and restore EGFP expression and was made using standard PMO synthesis methodology. The EEV were generated and conjugated to the EGFP-PMO essentially as disclosed in WO 2022/213118, entitled "CYCLIC CELL PENETRATING PEPTIDES", the disclosure of which is hereby incorporated by reference in its entirety.

Example 6. In Vitro Efficacy in HeLa654 Cells.

[0636] HeLa-EGFP-654 cells (LCCC Tissue Culture Facility, University of North Carolina at Chapel Hill, Chapel Hill, NC) were cultured in F-12K medium (ATCC) supplemented with 10% FBS and 400 µg/mL G418, in a humidified incubator at 37°C and 5% CO₂. Cells were plated at a density of 10⁴ cells/well in 96-well plates. After overnight culture, unconjugated PMO or EEV-PMO conjugate was added to the cells at the indicated concentrations to test for dose-dependent EGFP expression. The assay was performed in triplicate (n=3). Cells treated with vehicle (saline) only were used to obtain a blank measurement. At 24 h post treatment, the cells were washed and stained with Hoechst 33342. The mean fluorescence intensity of EGFP in the cells was quantified using a PerkinElmer Operetta CLS high content imager. Values are normalized to EGFP MFI signal produced by the unconjugated PMO and given as the fold-increase in signal over unconjugated PMO.

Table 3. EGFP expression in HeLa654 cells

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 µM, 24h)
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-K(Ac-Bip)-CO-PMO	4.9
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	4.8
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-CO-PMO	4.1
Ac-KKK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RK-Nal-CO-PMO	1.7
Ac-KK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-KRK-Nal-CO-PMO	1.7
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	2.1
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-CO-PMO	2.2

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 μ M, 24h)
Ac-K(<i>cyclo</i> [FGFGRGRQ])-KKKRRK-PEG ₁₂ -Nal-CO-PMO	2.7
Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-RK-PEG ₁₂ -Nal-CO-PMO	2.1
Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	4.1
Ac-KKK-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -rk-nal-CO-PMO	1.9
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkrk(Ac-nal)-CO-PMO	2.3
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -kkkr-nal-CO-PMO	1.7
Ac-PKKKRRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -Nal-CO-PMO	9.8
Ac-PKKKRRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRGQ])-PEG ₁₂ -Nal-CO-PMO	5
Ac-PKKKRRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Nal-PEG ₂ -Nal-CO-PMO	7.8
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -dNal-CO-PMO	132
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -Nal-CO-PMO	156
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fffrrrrQ])-PEG ₁₂ -dNal-CO-PMO	182
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -dNal-CO-PMO	150
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -dNal-CO-PMO	18
Ac-PKKKRRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-CO-PMO	11.1
Ac-KKKRRK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-CO-PMO	33.5
Ac-PKKKRRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-CO-PMO	27.4
Ac-PKKKRRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-CO-PMO	27.8
Ac-PKKKRRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-CO-PMO	38.7
Ac-PKKKRRKG-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-CO-PMO	11.4
<i>cyclo</i> [FGFGRGRQ]-Lys(Bip-KRKKKP-Ac)-CO-PMO	9.2
<i>cyclo</i> [FGFGRGRQ]-Lys(PKKKRRK-Bip-Ac)-CO-PMO	20.3
Ac-KKKRRK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Nal-CO-PMO	10.7
Ac-PKKKRRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Nal-CO-PMO	4

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 μ M, 24h)
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-Nal-CO-PMO	10
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-Nal-CO-PMO	21.3
Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG12-Nal-CO-PMO	7.9
Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG12-Nal-CO-PMO	23.2
Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG12-Nal-CO-PMO	19.1
Ac-RBRRBR-PEG2-K(cyclo[FfFGRGRQ])-PEG12-Nal-CO-PMO	66
Ac-RBRRBR-PEG2-K(cyclo[FfFRRRRQ])-PEG12-Nal-CO-PMO	171.4
Ac-RBRRBR-PEG2-K(cyclo[FfFRrRrQ])-PEG12-Nal-CO-PMO	135
Ac-rbrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-CO-PMO	15.3

Example 7. Ex Vivo Tolerability in Primary Human Proximal Tubule Epithelial Cells.

[0637] Primary Human Proximal Tubule Epithelial Cells (hRPTEC) were cultured according to the manufacturers protocol. Cells were harvested and plated at a density of 10^4 cells in opaque white 96-well plates. After overnight culture, a serial dilution of EEV-PMOs conjugate were added to the cells in saline and incubated. After 24 h, cell viability was determined using CellTiter Glo 2.0 and luminescence was measured using a SpectraMAX ID5 spectrophotometer and IC₅₀ values were determined using GraphPad Prism.

Table 4. hRPTEC Viability

Sequence	hRPTEC Viability IC ₅₀ , 24 h (μ M)
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-K(Ac-Bip)-CO-PMO	244.6
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	441.7
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-CO-PMO	255
Ac-KKK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RK-Nal-CO-PMO	531.3

Sequence	hRPTEC Viability IC ₅₀ , 24 h (μM)
Ac-KK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-KRK-Nal-CO-PMO	428.6
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	491.7
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	298
Ac-K(cyclo[FGFGRGRQ])-KKRK-PEG ₁₂ -Nal-CO-PMO	48.44
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-CO-PMO	87.13
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	142.7
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-CO-PMO	417.6
Ac-K(cyclo[FGFGRGRQ])-PEG12-kkkrk(Ac-nal)-CO-PMO	814.5
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-CO-PMO	142.7
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₁₂ -Nal-CO-PMO	51.71
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRGQ])-PEG ₁₂ -Nal-CO-PMO	84.13
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-PEG ₂ -Nal-CO-PMO	76.71
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-dNal-CO-PMO	48.9
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-Nal-CO-PMO	40
Ac-pkkkrkv-PEG2-k(cyclo[fffrrrrQ])-PEG12-dNal-CO-PMO	4.5
Ac-pkkkrkv-PEG2-k(cyclo[FGFRRRRQ])-PEG12-dNal-CO-PMO	47.6
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-dNal-CO-PMO	623.8
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-BRBRB-CO-PMO	239.9
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-BRBRB-CO-PMO	168.3
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-RBRB-CO-PMO	205.3
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RB-Bip-RB-CO-PMO	234.1
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RBRB-Bip-CO-PMO	194.2
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-BRBR-Bip-CO-PMO	266.8

Sequence	hRPTEC Viability IC ₅₀ , 24 h (μM)
cyclo[FGFGRGRQ]-Lys(Bip-KRKKKP-Ac)-CO-PMO	63.59
cyclo[FGFGRGRQ]-Lys(PKKKRK-Bip-Ac)-CO-PMO	25.02
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	296.5
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	290.4
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-Nal-CO-PMO	213.1
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-Nal-CO-PMO	99.12
Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG12-Nal-CO-PMO	100.9
Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG12-Nal-CO-PMO	12.22
Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG12-Nal-CO-PMO	47.57
Ac-RBRRBR-PEG2-K(cyclo[FfFGRGRQ])-PEG12-Nal-CO-PMO	36.3
Ac-RBRRBR-PEG2-K(cyclo[FfFRRRRQ])-PEG12-Nal-CO-PMO	16.9
Ac-RBRRBR-PEG2-K(cyclo[FfFRrRrQ])-PEG12-Nal-CO-PMO	2.2
Ac-rbrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-CO-PMO	95.8

Example 8. In Vivo Efficacy in EGFP654 Mice.

[0638] EEV-PMO conjugates were administered to EGFP654 mice at a dose of 5 mg/kg injection in a volume of 5 mL/kg of sterile saline. After 48 h, animals were sacrificed and tissues were harvested, snap-frozen and pulverized. To determine splice correction of EGFP transcripts, RNA was extracted from pulverized tissues using the RNeasy Mini Kit (QIAGEN, Cat No./ID: 74106, Hilden, Germany) per the manufacturer's protocol. RT-PCR was performed using 200 ng of the extracted RNA and the QIAGEN OneStep RT-PCR Kit (QIAGEN) per manufacturer's instructions. The reverse transcription was carried out at 50 °C for 30 min. Then the PCR reaction was carried out with the following program: 1 cycle, 94 °C, 15 min; 35 cycles, 94 °C, 30 s; 60 °C, 30 s; 72 °C, 30 s; 1 cycle, 72 °C, 10 min. The forward and reverse primers were 5'-CGTAAACGGCCACAAGTTCAGCG -3' and 5'-GTGGTGCAGATGAACTTCAGGGTC -3', respectively. PCR products were analyzed by 2% E-gel (Thermo Fisher Scientific). Product bands were quantified by densitometry analysis with ImageJ software, and skipping efficacy was

calculated with the following equation: skipping (%) = $A/(A+B) \times 100$, with A = the intensity of the skipped band and B = the intensity of the full-length band.

Table 5. Splice Correction

Sequence	Gastroc Splice Correction, 5 mpk, 48 h PI	Heart Splice Correction, 5 mpk, 48 h PI
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	9.9	0
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG12-Bip-CO-PMO	15.5	0.9
Ac-KKK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG12-RK-Nal-CO-PMO	0	0
Ac-KK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG12-KRK-Nal-CO-PMO	6.6	0
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	0	0
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	3.2	0
Ac-K(cyclo[FGFGRGRQ])-KKRK-PEG ₁₂ -Nal-CO-PMO	38.3	22.2
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-CO-PMO	23.1	3.9
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	25.6	0
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-CO-PMO	4.3	0
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-CO-PMO	0	0
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₁₂ -Nal-CO-PMO	45.7	42
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRGQ])-PEG ₁₂ -Nal-CO-PMO	32.7	18.8
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-PEG ₂ -Nal-CO-PMO	61.8	58.1
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG12-dNal-CO-PMO	15.1	16.8
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG12-Nal-CO-PMO	22.7	30.9
Ac-pkkkrkv-PEG ₂ -k(cyclo[fffrrrQ])-PEG12-dNal-CO-PMO	30.8	40

Ac-pkkkrkv-PEG2-k(cyclo[FGFRRRRQ])-PEG12-dNal-CO-PMO	25.1	25.9
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-dNal-CO-PMO	25.8	2.8
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-BRBRB-CO-PMO	11.9	9.5
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-BRBRB-CO-PMO	16.4	10.6
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-RBRB-CO-PMO	7.2	5.6
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RB-Bip-RB-CO-PMO	5.3	0
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-BHRHR-CO-PMO	0	0.9
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-BRHRH-CO-PMO	1.7	0.2
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RHRH-Bip-CO-PMO	6.6	0
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RBRB-Bip-CO-PMO	7.2	2.2
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-BRBR-Bip-CO-PMO	0	3
Ac-PKKKRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-HRHR-Bip-CO-PMO	4.7	0
cyclo[FGFGRGRQ]-Lys(Bip-KRKKKP-Ac)-CO-PMO	0	0
cyclo[FGFGRGRQ]-Lys(PKKKRK-Bip-Ac)-CO-PMO	0	0
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	43.7	9.4
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	29.9	3
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-Nal-CO-PMO	45.8	35.3
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-Nal-CO-PMO	44.5	23.1
Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG12-Nal-CO-PMO	11.6	0
Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG12-Nal-CO-PMO	57.3	45.2

Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG12-Nal-CO-PMO	26.5	34.9
Ac-RBRRBR-PEG2-K(cyclo[FfFGRGRQ])-PEG12-Nal-CO-PMO	62.1	6.1
Ac-RBRRBR-PEG2-K(cyclo[FfFRRRRQ])-PEG12-Nal-CO-PMO	39.4	30.4
Ac-RBRRBR-PEG2-K(cyclo[FfFRrRrQ])-PEG12-Nal-CO-PMO	54.5	48.6
Ac-rbrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-CO-PMO	12.8	0

Example 9. In Vivo Tolerability in CD1 Mice.

[0639] EEV-PMO conjugates were administered intravenously to CD1 mice at a dose of 50 mg/kg formulated in 5 mL/kg saline. After injection, mice were assessed for in-life observations relating to time to return to baseline. The scores were assigned as follows: 0 = baseline, 1 = < 1 h to recovery, 2 = < 4 h to recovery, 3 = < 24 h to recovery.

Table 6. In Vivo Tolerability

Sequence	In-Life Score, CD1, 50 mpk
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-K(Ac-Bip)-CO-PMO	1
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Nal-CO-PMO	1
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG12-Bip-CO-PMO	1
Ac-KK-PEG2-K(cyclo[FGFGRGRQ])-PEG12-KRK-Nal-CO-PMO	1
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	1
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	1
Ac-K(cyclo[FGFGRGRQ])-KKRK-PEG ₁₂ -Nal-CO-PMO	3
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-CO-PMO	2
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	1
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-CO-PMO	1
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-CO-PMO	1
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₁₂ -Nal-CO-PMO	3

Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFRRRGQ])-PEG ₁₂ -Nal-CO-PMO	3
Ac-PKKKRKV-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -Nal-PEG ₂ -Nal-CO-PMO	3
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -dNal-CO-PMO	3
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfrrrrQ])-PEG ₁₂ -Nal-CO-PMO	3
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fffrrrrQ])-PEG ₁₂ -dNal-CO-PMO	3
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -dNal-CO-PMO	3
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -dNal-CO-PMO	1

Example 10. Ex Vivo Tolerability in Primary Human Proximal Tubule Epithelial Cells.

[0640] Primary Human Proximal Tubule Epithelial Cells (hRPTEC) were cultured according to the manufacturers protocol. Cells were harvested and plated at a density of 10^4 cells in opaque white 96-well plates. After overnight culture, a serial dilution of EEV-PMOs conjugate were added to the cells in saline and incubated. After 24 h, cell viability was determined using CellTiter Glo 2.0 and luminescence was measured using a SpectraMAX ID5 spectrophotometer and IC₅₀ values were determined using GraphPad Prism.

Table 7. hRPTEC Viability

Sequence	hRPTEC Viability IC ₅₀ , 24 h (μM)
Ac-KKK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	531.3
Ac-KK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKK-Nal-CO-PMO	428.6
Ac-K-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	491.7
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-CO-PMO	298

Example 11. In Vivo Efficacy in EGFP654 Mice.

[0641] EEV-PMO conjugates were administered to EGFP654 mice at a dose of 5 mg/kg injection in a volume of 5 mL/kg of sterile saline. After 48 h, animals were sacrificed and tissues were harvested, snap-frozen and pulverized. To determine splice correction of EGFP transcripts, RNA was extracted from pulverized tissues using the RNeasy Mini Kit (QIAGEN, Cat No./ID: 74106,

Hilden, Germany) per the manufacturer's protocol. RT-PCR was performed using 200 ng of the extracted RNA and the QIAGEN OneStep RT-PCR Kit (QIAGEN) per manufacturer's instructions. The reverse transcription was carried out at 50 °C for 30 min. Then the PCR reaction was carried out with the following program: 1 cycle, 94 °C, 15 min; 35 cycles, 94 °C, 30 s; 60 °C, 30 s; 72 °C, 30 s; 1 cycle, 72 °C, 10 min. The forward and reverse primers were 5'-CGTAAACGGCCACAAGTTCAGCG -3' and 5'- GTGGTGCAGATGAACTTCAGGGTC -3', respectively. PCR products were analyzed by 2% E-gel (Thermo Fisher Scientific). Product bands were quantified by densitometry analysis with ImageJ software, and skipping efficacy was calculated with the following equation: skipping (%) = $A/(A+B) \times 100$, with A = the intensity of the skipped band and B = the intensity of the full-length band.

Table 8. Splice Correction

Sequence	Gastroc Splice Correction, 5 mpk, 48 h PI
Ac-KKK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	0
Ac-KK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKK-Nal-CO-PMO	6.6
Ac-K-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	0
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-CO-PMO	3.2

Example 12. In Vivo Tolerability in CD1 Mice.

[0642] EEV-PMO conjugates were administered intravenously to CD1 mice at a dose of 50 mg/kg formulated in 5 mL/kg saline. After injection, mice were assessed for in-life observations relating to time to return to baseline. The scores were assigned as follows: 0 = baseline, 1 = < 1 h to recovery, 2 = < 4 h to recovery, 3 = < 24 h to recovery.

Table 9. In vivo Tolerability

Sequence	In-Life Score, CD1, 50 mpk
Ac-KKK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -RK-Nal-CO-PMO	1

Ac-KK-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKK-Nal-CO-PMO	2
Ac-K-PEG ₂ -K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	1
Ac-K(<i>cyclo</i> [FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-CO-PMO	1

Example 13. In Vitro Efficacy in HeLa654 Cells.

[0643] In Vitro Efficacy was tested in HeLa654 Cells essentially as described in Example 6. EEV sequences are provided in Table 11 below and were prepared as described in Example 5. The PMO sequence is provided in Example 5.

Table 10. EGFP expression in HeLa654 cells

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 μ M, 24h)
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -dNal-CO-PMO	150.66
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -dNal-CO-PMO	18.08
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -CO-PMO	8.73

Example 14. Ex Vivo Tolerability in Primary Human Proximal Tubule Epithelial Cells.

[0644] Ex vivo tolerability was tested in Primary Human Proximal Tubule Epithelial Cells as described in Example 7. EEV sequences are provided in Table 12 below and were prepared as described in Example 5. The PMO sequence is provided in Example 5.

Table 11. hRPTEC Viability

Sequence	hRPTEC Viability IC ₅₀ , 24 h (μ M)
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [FGFRRRRQ])-PEG ₁₂ -dNal-CO-PMO	47.55
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -dNal-CO-PMO	623.8
Ac-pkkkrkv-PEG ₂ -k(<i>cyclo</i> [fGfGrGrQ])-PEG ₁₂ -CO-PMO	673.8

Example 15. In Vivo Efficacy in EGFP654 Mice.

[0645] In vivo efficacy was tested in EGFP654 Mice as described in Example 8. EEV sequences are provided in Table 13 below and were prepared as described in Example 5. The PMO sequence is provided in Example 5.

5 **Table 12. In Vivo Tolerability**

Sequence	Gastroc Splice Correction, 5 mpk, 48 h PI
Ac-pkkkrkv-PEG ₂ -k(cyclo[FGFRRRRQ])-PEG ₁₂ -dNal-CO-PMO	25.1
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -dNal-CO-PMO	25.8
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -CO-PMO	0

Example 16. In Vivo Tolerability in CD1 Mice.

[0646] In vivo tolerability was tested in CD1 Mice as described above. EEV sequences are provided in Table 13 below and were prepared as described above. The PMO sequence is provided in Example 5.

10 **Table 13. In Vivo Tolerability**

Sequence	In-Life Score, CD1, 50 mpk
Ac-pkkkrkv-PEG ₂ -k(cyclo[FGFRRRRQ])-PEG ₁₂ -dNal-CO-PMO	3
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -dNal-CO-PMO	1
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -CO-PMO	0

Example 17. In Vitro Efficacy in HeLa654 Cells.

[0647] HeLa-EGFP-654 cells (LCCC Tissue Culture Facility, University of North Carolina at Chapel Hill, Chapel Hill, NC) were cultured in F-12K medium (ATCC) supplemented with 10% FBS and 400 µg/mL G418, in a humidified incubator at 37°C and 5% CO₂. Cells were plated at a density of 104 cells/well in 96-well plates. After overnight culture, unconjugated PMO or EEV-PMO conjugate was added to the cells at the indicated concentrations to test for dose-dependent

EGFP expression. The assay was performed in triplicate (n=3). Cells treated with vehicle (saline) only were used to obtain a blank measurement. At 24 h post treatment, the cells were washed and stained with Hoechst 33342. The mean fluorescence intensity of EGFP in the cells was quantified using a PerkinElmer Operetta CLS high content imager. Values are normalized to EGFP MFI signal produced by the unconjugated PMO and given as the fold-increase in signal over unconjugated PMO.

Table 14A. EGFP expression in HeLa654 cells

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 μM, 24h)
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILYBRBRB-OH	267.8
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILY-PEG2-OH	216.2
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-OH	42
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	58.5
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-Bip-OH	207.7
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG2-Bip-OH	253.6
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-K(N3)-NH2	30
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	74
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-R-Bip-OH	12.7
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF-OH	14.3
Ac-KKKRK-PEG2-K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	209.2
Ac-KKKRK-PEG2-K(RBRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	158.8

Ac-RBRRBR-PEG2-K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	207.5
Ac-KRK-PEG2-K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	12.6
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-RF-PEG4-OH	7
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF-PEG4-OH	6
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-RF-PEG4-OH	7.7
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-B-RF-PEG4-OH	8.6
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG4-RF-PEG4-OH	16.4

Table 14B. EGFP expression in HeLa654 cells

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 μ M, 24h)
K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^u MeK-N ^u MeK-N ^u MeK-N ^u MeR-N ^u MeK(Ac-Nal)-OH	4.4
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^u MeK-N ^u MeK-N ^u MeK-N ^u MeR-N ^u MeK(Ac-nal)-OH	4.6
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^u MeK-N ^u MeK-N ^u MeR-N ^u MeK(Ac-Nal)-OH	4.4
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^u MeK-N ^u MeK-N ^u MeR-N ^u MeK(Ac-nal)-OH	3.4
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^u MeK-N ^u MeK-N ^u MeK-N ^u MeR-k(N ₃)-NH ₂	5.4
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^u MeK-N ^u MeK-N ^u MeR-k(N ₃)-NH ₂	4.8

Example 18. Ex Vivo Tolerability in Primary Human Proximal Tubule Epithelial Cells.

[0648] Primary Human Proximal Tubule Epithelial Cells (hRPTEC) were cultured according to the manufacturers protocol. Cells were harvested and plated at a density of 104 cells in opaque white 96-well plates. After overnight culture, a serial dilution of EEV-PMOs conjugate were added to the cells in saline and incubated. After 24 h, cell viability was determined using CellTiter Glo 2.0 and luminescence was measured using a SpectraMAX ID5 spectrophotometer and IC50 values were determined using GraphPad Prism.

Table 15A. hRPTEC Viability

Sequence	hRPTEC Viability IC50, 24 h (μM)
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILYBRBRB-OH	18.17
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILY-PEG2-OH	17.2
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-OH	61.81
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	89.8
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-Bip-OH	23.37
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG2-Bip-OH	35.56
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-K(N3)-NH2	39.9
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	70.55
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-R-Bip-OH	112.2
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF-OH	347.4
Ac-KKKRK-PEG2-K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	19.94

Ac-KKKRK-PEG2-K(RBRRBR-Ac)- K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	19.68
Ac-RBRRBR-PEG2-K(KKRKK-Ac)- K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	17.75
Ac-KRK-PEG2-K(KRK-Ac)-K(cyclo[FGFGRGRQ])- PEG12-K(N3)-NH2	112.8
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-RF- PEG4-OH	226.5
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF- PEG4-OH	278
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-RF-PEG4- OH	244.7
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-B-RF- PEG4-OH	165.7
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG4-RF- PEG4-OH	151.6

Table 15B. hRPTEC Viability

Sequence	hRPTEC Viability IC ₅₀ , 24 h (μ M)
K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{Me} K-N ^{Me} K-N ^{Me} K- N ^{Me} R-N ^{Me} K(Ac-Nal)-OH	439.1
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{Me} K-N ^{Me} K-N ^{Me} K- N ^{Me} R-N ^{Me} K(Ac-nal)-OH	462.4
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{Me} K-N ^{Me} K-N ^{Me} R- N ^{Me} K(Ac-Nal)-OH	395.2
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{Me} K-N ^{Me} K-N ^{Me} R- N ^{Me} K(Ac-nal)-OH	381.3
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -NMeK-NMeK- NMeR-Nal-OH	983.1
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{Me} K-N ^{Me} K-N ^{Me} K- N ^{Me} R-k(N ₃)-NH ₂	240.5
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -N ^{Me} K-N ^{Me} K-N ^{Me} R- k(N ₃)-NH ₂	283.3

Ac-K(cyclo[FGFGRGRQ])-PEG12-NMeK-NMeR-OH	832.1
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Example 19. In Vivo Efficacy in EGFP654 Mice.

[0649] EEV-PMO conjugates were administered to EGFP654 mice at a dose of 5 mg/kg injection in a volume of 5 mL/kg of sterile saline. After 48 h, animals were sacrificed and tissues were harvested, snap-frozen and pulverized. To determine splice correction of EGFP transcripts, RNA was extracted from pulverized tissues using the RNeasy Mini Kit (QIAGEN, Cat No./ID: 74106, Hilden, Germany) per the manufacturer's protocol. RT-PCR was performed using 200 ng of the extracted RNA and the QIAGEN OneStep RT-PCR Kit (QIAGEN) per manufacturer's instructions. The reverse transcription was carried out at 50 °C for 30 min. Then the PCR reaction was carried out with the following program: 1 cycle, 94 °C, 15 min; 35 cycles, 94 °C, 30 s; 60 °C, 30 s; 72 °C, 30 s; 1 cycle, 72 °C, 10 min. The forward and reverse primers were 5'-CGTAAACGGCCACAAGTTCAGCG -3' and 5'-GTGGTGCAGATGAACTTCAGGGTC -3', respectively. PCR products were analyzed by 2% E-gel (Thermo Fisher Scientific). Product bands were quantified by densitometry analysis with ImageJ software, and skipping efficacy was calculated with the following equation: skipping (%) = $A/(A+B) \times 100$, with A = the intensity of the skipped band and B = the intensity of the full-length band.

Table 16. Splice Correction

Sequence	Gastroc Splice Correction, 5 mpk, 48 h PI	Heart Splice Correction, 5 mpk, 48 h PI
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILYBRBRB-OH	35.1	52.4
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILY-PEG2-OH	80.7	79.8
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-OH	63.3	78.3
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	66.5	78.4
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-Bip-OH	59.4	65.4

Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG2-Bip-OH	59.2	51.1
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-K(N3)-NH2	79.9	83.6
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	84.8	86.4
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-R-Bip-OH	52.6	49.6
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF-OH	27.3	20.5
Ac-KKKRK-PEG2-K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	1.26	0
Ac-KKKRK-PEG2-K(RBRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	0	0
Ac-RBRRBR-PEG2-K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	0	0
Ac-KRK-PEG2-K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG12-K(N3)-NH2	10.47	0
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-RF-PEG4-OH	84	89.6
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG4-RF-PEG4-OH	89.2	91.1
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG8-RF-PEG4-OH	80.3	83.3
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-OH	80.8	72.9
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG4-RF-PEG4-OH	22.4	0
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF-PEG4-OH	13.7	0
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-RF-PEG4-OH	65.7	47.2
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-B-RF-PEG4-OH	52.6	21.1
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG4-RF-PEG4-OH	44.5	18.4
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG8-RF-PEG4-OH	39.3	17.5

Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-R-PEG4-OH	1.9	0
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-F-PEG4-OH	24.7	2.2
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RRF-PEG4-OH	68.7	69.2
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RFR-PEG4-OH	84.2	87.9

Example 20. In Vivo Tolerability in CD1 Mice.

[0650] EEV-PMO conjugates were administered intravenously to CD1 mice at a dose of 50 mg/kg formulated in 5 mL/kg saline. After injection, mice were assessed for in-life observations relating to time to return to baseline. The scores were assigned as follows: 0 = baseline, 1 = < 1 h to recovery, 2 = < 4 h to recovery, 3 = < 24 h to recovery.

Table 17A. In Vivo Tolerability

Sequence	In-Life Score, CD1, 50 mpk
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILYBRBRB-OH	3
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RFQILY-PEG2-OH	3
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-OH	2
Ac-PKKKRKV-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	2
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG4-Bip-OH	3
Ac-PKKKRKV-PEG2-K(cyclo[FfFGRGRQ])-PEG2-RF-PEG2-Bip-OH	2
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-K(N3)-NH2	3
Ac-KKKRK-PEG2-K(cyclo[FGFGRGRQ])-PEG2-RF-PEG4-Bip-OH	2

Example 21. In Vitro Efficacy in HeLa654 Cells.

[0651] HeLa-EGFP-654 cells (LCCC Tissue Culture Facility, University of North Carolina at Chapel Hill, Chapel Hill, NC) were cultured in F-12K medium (ATCC) supplemented with 10% FBS and 400 µg/mL G418, in a humidified incubator at 37°C and 5% CO₂. Cells were plated at

a density of 10^4 cells/well in 96-well plates. After overnight culture, unconjugated PMO or EEV-PMO conjugate was added to the cells at the indicated concentrations to test for dose-dependent EGFP expression. The assay was performed in triplicate ($n=3$). Cells treated with vehicle (saline) only were used to obtain a blank measurement. At 24 h post treatment, the cells were washed and stained with Hoechst 33342. The mean fluorescence intensity of EGFP in the cells was quantified using a PerkinElmer Operetta CLS high content imager. Values are normalized to EGFP MFI signal produced by the unconjugated PMO and given as the fold-increase in signal over unconjugated PMO.

Table 18. EEV-PMOs

Sequence	HeLa654, MFI Fold Increase over PMO @ 30 μ M, 24h)
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-dNal-CO-PMO	132
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-Nal-CO-PMO	156
Ac-pkkkrkv-PEG2-k(cyclo[fffrrrrQ])-PEG12-dNal-CO-PMO	182
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-dNal-CO-PMO	18
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-CO-PMO	9
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-K(N3)-NH2	15.1
Ac-rbrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-CO-PMO	15.3

Example 22. Ex Vivo Tolerability in Primary Human Proximal Tubule Epithelial Cells.

[0652] Primary Human Proximal Tubule Epithelial Cells (hRPTEC) were cultured according to the manufacturers protocol. Cells were harvested and plated at a density of 10^4 cells in opaque white 96-well plates. After overnight culture, a serial dilution of EEV-PMOs conjugate were added to the cells in saline and incubated. After 24 h, cell viability was determined using CellTiter Glo 2.0 and luminescence was measured using a SpectraMAX ID5 spectrophotometer and IC_{50} values were determined using GraphPad Prism.

Table 19. hRPTEC Viability

Sequence	hRPTEC Viability IC_{50} , 24 h (μ M)
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-dNal-CO-PMO	48.9
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-Nal-CO-PMO	40
Ac-pkkkrkv-PEG2-k(cyclo[fffrrrrQ])-PEG12-dNal-CO-PMO	4.5
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-dNal-CO-PMO	623.8

Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-CO-PMO	673.8
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-K(N3)-NH2	320.3
Ac-rbrrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-CO-PMO	95.8

Example 23. In Vivo Efficacy in EGFP654 Mice.

[0653] EEV-PMO conjugates were administered to EGFP654 mice at a dose of 5 mg/kg injection in a volume of 5 mL/kg of sterile saline. After 48 h, animals were sacrificed and tissues were harvested, snap-frozen and pulverized. To determine splice correction of EGFP transcripts, RNA was extracted from pulverized tissues using the RNeasy Mini Kit (QIAGEN, Cat No./ID: 74106, Hilden, Germany) per the manufacturer's protocol. RT-PCR was performed using 200 ng of the extracted RNA and the QIAGEN OneStep RT-PCR Kit (QIAGEN) per manufacturer's instructions. The reverse transcription was carried out at 50 °C for 30 min. Then the PCR reaction was carried out with the following program: 1 cycle, 94 °C, 15 min; 35 cycles, 94 °C, 30 s; 60 °C, 30 s; 72 °C, 30 s; 1 cycle, 72 °C, 10 min. The forward and reverse primers were 5'-CGTAAACGGCCACAAGTTCAGCG -3' and 5'-GTGGTGCAGATGAACTTCAGGGTC -3', respectively. PCR products were analyzed by 2% E-gel (Thermo Fisher Scientific). Product bands were quantified by densitometry analysis with ImageJ software, and skipping efficacy was calculated with the following equation: skipping (%) = $A/(A+B) \times 100$, with A = the intensity of the skipped band and B = the intensity of the full-length band.

Table 20. Splice Correction

Sequence	Gastroc Splice Correction, 5 mpk, 48 h PI	Heart Splice Correction, 5 mpk, 48 h PI
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-dNal-CO-PMO	15.1	16.8
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-Nal-CO-PMO	22.7	30.9
Ac-pkkkrkv-PEG2-k(cyclo[ffffrrrrQ])-PEG12-dNal-CO-PMO	30.8	40
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-dNal-CO-PMO	25.8	2.8
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-CO-PMO	0	0
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-K(N3)-NH2	29.4	0

Ac-rbrrbr-PEG2-K(cyclo[fGfGrGrQ])-PEG12-Nal-CO-PMO	12.8	0
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Example 24. In Vivo Tolerability in CD1 Mice.

[0654] EEV-PMO conjugates were administered intravenously to CD1 mice at a dose of 50 mg/kg formulated in 5 mL/kg saline. After injection, mice were assessed for in-life observations relating to time to return to baseline. The scores were assigned as follows: 0 = baseline, 1 = < 1 h to recovery, 2 = < 4 h to recovery, 3 = < 24 h to recovery.

Table 21. EEV-PMOs

Sequence	In-Life Score, CD1, 50 mpk
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-dNal-CO-PMO	3
Ac-pkkkrkv-PEG2-k(cyclo[fGfrrrrQ])-PEG12-Nal-CO-PMO	3
Ac-pkkkrkv-PEG2-k(cyclo[fffrrrrQ])-PEG12-dNal-CO-PMO	3
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-dNal-CO-PMO	1
Ac-pkkkrkv-PEG2-k(cyclo[fGfGrGrQ])-PEG12-CO-PMO	0

Example 25. In Vitro Efficacy in HeLa654 Cells.

[0655] HeLa-EGFP-654 cells (LCCC Tissue Culture Facility, University of North Carolina at Chapel Hill, Chapel Hill, NC) were cultured in F-12K medium (ATCC) supplemented with 10% FBS and 400 µg/mL G418, in a humidified incubator at 37°C and 5% CO₂. Cells were plated at a density of 104 cells/well in 96-well plates. After overnight culture, unconjugated PMO or EEV-PMO conjugate was added to the cells at the indicated concentrations to test for dose-dependent EGFP expression. The assay was performed in triplicate (n=3). Cells treated with vehicle (saline) only were used to obtain a blank measurement. At 24 h post treatment, the cells were washed and stained with Hoechst 33342. The mean fluorescence intensity of EGFP in the cells was quantified using a PerkinElmer Operetta CLS high content imager. Values are normalized to EGFP MFI signal produced by the unconjugated PMO and given as the fold-increase in signal over unconjugated PMO.

Table 22. EGFP expression in HeLa654 cells

Sequence	HeLa654, MFI Fold
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	Increase over PMO @ 30 μ M, 24h)
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH	11.1
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH	33.5
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH	27.4
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-OH	27.8
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH	38.7
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH	11.4
cyclo[FGFGRGRQ]-K(Bip-KRKKKP-Ac)-OH	9.2
cyclo[FGFGRGRQ]-K(PKKKRK-Bip-Ac)-OH	20.3

Example 26. Ex Vivo Tolerability in Primary Human Proximal Tubule Epithelial Cells.

[0656] Primary Human Proximal Tubule Epithelial Cells (hRPTEC) were cultured according to the manufacturers protocol. Cells were harvested and plated at a density of 104 cells in opaque white 96-well plates. After overnight culture, a serial dilution of EEV-PMOs conjugate were added to the cells in saline and incubated. After 24 h, cell viability was determined using CellTiter Glo 2.0 and luminescence was measured using a SpectraMAX ID5 spectrophotometer and IC50 values were determined using GraphPad Prism.

Table 23. hRPTEC Viability

Sequence	hRPTEC Viability IC50, 24 h (μ M)
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH	239.9
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH	168.3
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH	205.3
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-OH	234.1
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH	194.2
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH	266.8
cyclo[FGFGRGRQ]-K(Bip-KRKKKP-Ac)-OH	63.59

cyclo[FGFGRGRQ]-K(PKKKRK-Bip-Ac)-OH	25.02
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Example 27. In Vivo Efficacy in EGFP654 Mice.

[0657] EEV-PMO conjugates were administered to EGFP654 mice at a dose of 5 mg/kg injection in a volume of 5 mL/kg of sterile saline. After 48 h, animals were sacrificed and tissues were harvested, snap-frozen and pulverized. To determine splice correction of EGFP transcripts, RNA was extracted from pulverized tissues using the RNeasy Mini Kit (QIAGEN, Cat No./ID: 74106, Hilden, Germany) per the manufacturer's protocol. RT-PCR was performed using 200 ng of the extracted RNA and the QIAGEN OneStep RT-PCR Kit (QIAGEN) per manufacturer's instructions. The reverse transcription was carried out at 50 °C for 30 min. Then the PCR reaction was carried out with the following program: 1 cycle, 94 °C, 15 min; 35 cycles, 94 °C, 30 s; 60 °C, 30 s; 72 °C, 30 s; 1 cycle, 72 °C, 10 min. The forward and reverse primers were 5'-CGTAAACGGCCACAAGTTCAGCG -3' and 5'-GTGGTGCAGATGAACTTCAGGGTC -3', respectively. PCR products were analyzed by 2% E-gel (Thermo Fisher Scientific). Product bands were quantified by densitometry analysis with ImageJ software, and skipping efficacy was calculated with the following equation: skipping (%) = $A/(A+B) \times 100$, with A = the intensity of the skipped band and B = the intensity of the full-length band.

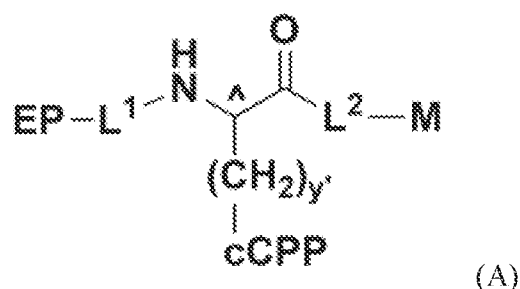
Table 24. Gastroc. Splice Correction

Sequence	Gastroc Splice Correction, 5 mpk, 48 h PI	Heart Splice Correction, 5 mpk, 48 h PI
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH	11.9	9.5
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH	16.4	10.6
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH	7.2	5.6
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RB-Bip-RB-OH	5.3	0
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BHRHR-OH	0	0.9
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRHRH-OH	1.7	0.2

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-OH	6.6	0
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH	7.2	2.2
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH	0	3
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-OH	4.7	0
cyclo[FGFGRGRQ]-K(Bip-KRKKKP-Ac)-OH	0	0
cyclo[FGFGRGRQ]-K(PKKKRK-Bip-Ac)-OH	0	0

CLAIMS

1. An endosomal escape vehicle (EEV) comprising the structure:



wherein

- 5 EP is a linear exocyclic peptide;
 cCPP is a cyclic cell penetrating peptide;
 L¹ and L², are, independently, a linker arm;
 ^ indicates L- or D-stereochemistry;
 y' is an integer from 1 to 5; and
 10 M comprises a reactive handle.
2. The EEV of claim 1, wherein L¹ comprises a polyethylene glycol component (PEG)x', an amino acid component (AA)a', , or a combination thereof, wherein a' is an integer from 0-12 and x' is an integer from 0-12.
- 15 3. The EEV of claim 1 or 2, wherein L¹ comprises (PEG)x' and x' is an integer from 1 to 12.
4. The EEV of claim 2, wherein x' is an integer from 0, 2, 4, 8, or 12.
- 20 5. The EEV of any of claims 2-4, wherein x' is 0 or 2.
6. The EEV of any of claims 2-5, wherein (AA)a' comprises one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β-alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof.
- 25 7. The EEV of claim 6, wherein (AA)a' comprises at least one charged amino acid.

8. The EEV of claim 6 or 7, wherein a' is an integer from 1-12.

9. The EEV of claim 6 or 7, wherein a' is an integer from 1-8.

5

10. The EEV of claim 6 or 7, wherein a' is an integer from 2-5.

11. The EEV of any of claims 1-10, wherein L^2 comprises one or more polyethylene glycol components $(\text{PEG})_{z'}$, $(\text{PEG})_{z''}$, one or more amino acid components $(\text{AA})_{b'}$, $(\text{AA})_{b''}$, one or
10 more hydrophobic components $(X^{o'})$, $(X^{o''})$, or a combination thereof, wherein b' and b'' are, independently, an integer from 0-12 and z' and z'' are, independently, an integer from 0-12.

12. The EEV of claim 11, wherein L^2 comprises $(\text{PEG})_{z'}$, $(\text{PEG})_{z''}$, or a combination thereof and z' and z'' are, independently, an integer from 1 to 12.

15

13. The EEV of claim 11, wherein L^2 comprises $(\text{PEG})_{z'}$, $(\text{PEG})_{z''}$, or a combination thereof and z' and z'' are, independently, 2, 4, 8, or 12.

14. The EEV of claim 11, wherein L^2 comprises $(\text{PEG})_{z'}$, and z' is 2, 4, 8, or 12.

20

15. The EEV of claim 11, wherein L^2 comprises $(\text{PEG})_{z'}$ and $(\text{PEG})_{z''}$, and z' and z'' are, independently, 2, 4, 8, or 12.

16. The EEV of any of claims 11-15, wherein $(\text{AA})_{b'}$ and $(\text{AA})_{b''}$ comprise one or more amino
25 acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof.

17. The EEV of claim 16, wherein $(\text{AA})_{b'}$ and $(\text{AA})_{b''}$ comprise at least one charged amino acid.

30

18. The EEV of claim 16 or 17, wherein b' and b'' are, independently, an integer from 1-12.

19. The EEV of claim 16 or 17, wherein b' and b'' are, independently, an integer from 1-8.

20. The EEV of claim 16 or 17, wherein b' and b'' are, independently, an integer from 2-5.

5

21. The EEV of any of claims 11-20, wherein L^2 comprises one or more hydrophobic components ($X^{o'}$), ($X^{o''}$).

22. The EEV of claim 21, wherein $X^{o'}$ and $X^{o''}$ are the same.

10

23. The EEV of claim 21, wherein $X^{o'}$ and $X^{o''}$ are different.

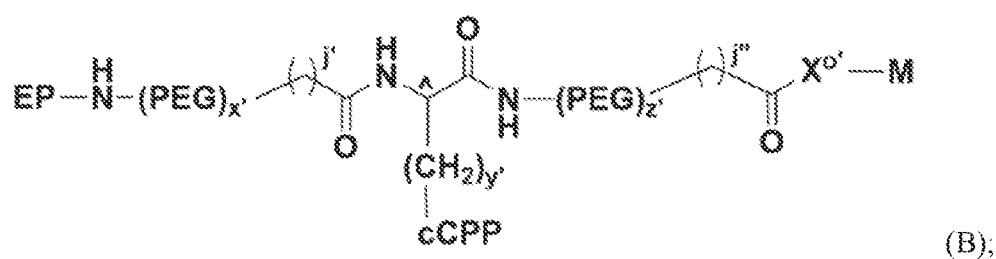
24. The EEV of claim 1, wherein L^1 , L^2 , cCPP or a combination thereof comprises a hydrophobic component (X).

15

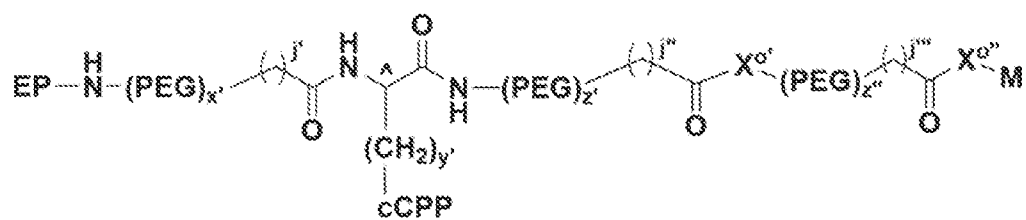
25. The EEV of any of claims 22-24, wherein the hydrophobic components ($X^{o'}$), ($X^{o''}$) are, independently, selected from Nal, dNal, or Bip.

26. The EEV of claim 1, comprising a structure selected from:

20 *Formula (B):*

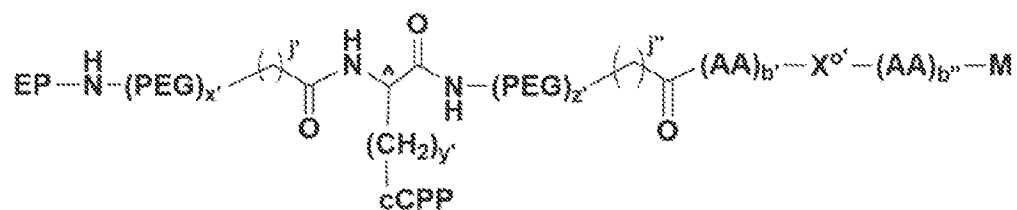


Formula (BB):



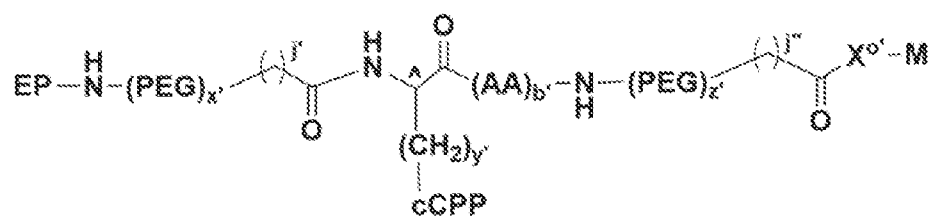
(BB); Formula

(C):



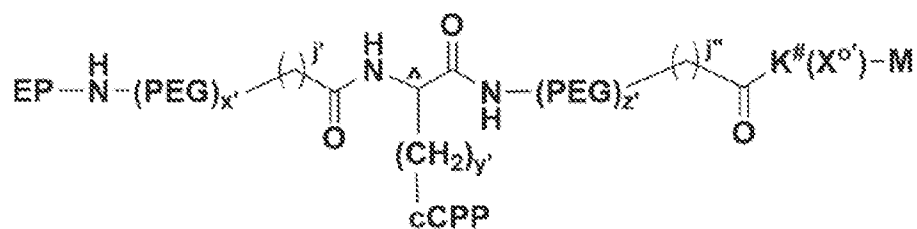
(C);

5 Formula (D):



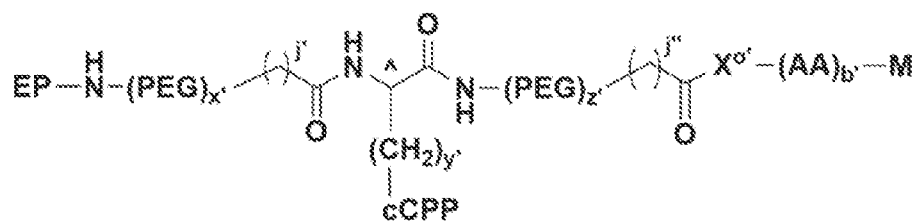
(D);

Formula (E):



(E); and

Formula (F):



(F);

10

wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

5 z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ and $X^{o''}$ each, individually, comprise a hydrophobic component;

$K^{\#}$ is D-lysine or L-lysine residue;

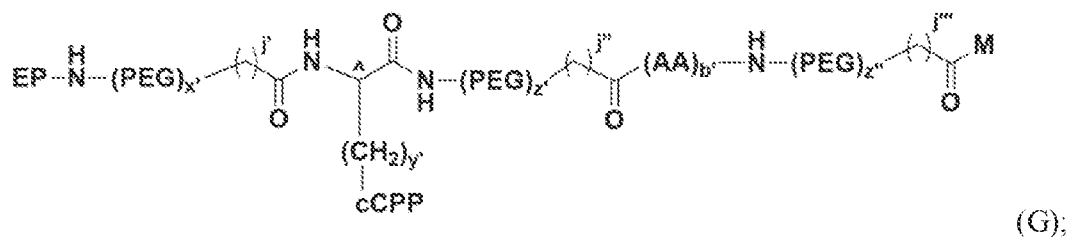
10 $(AA)b'$ and $(AA)b''$ are each an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

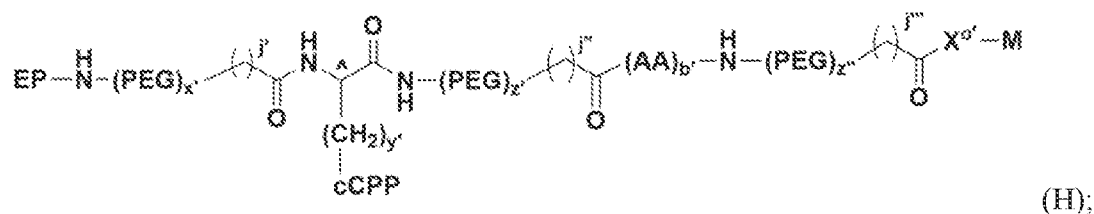
b'' is an integer from 0 to 12.

27. The EEV of claim 1, comprising a structure selected from:

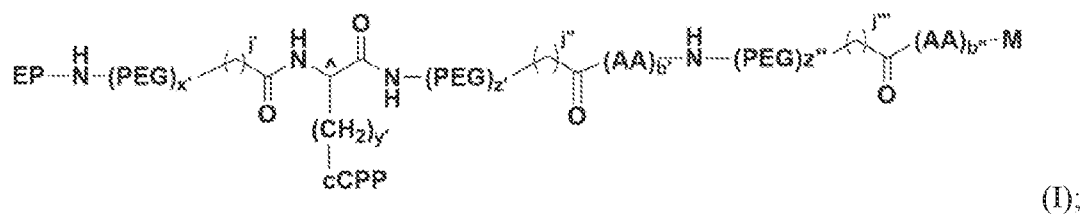
15 *Formula (G):*



Formula (H):

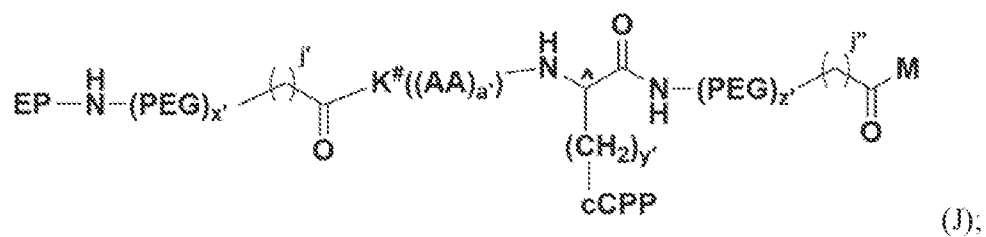


Formula (I):

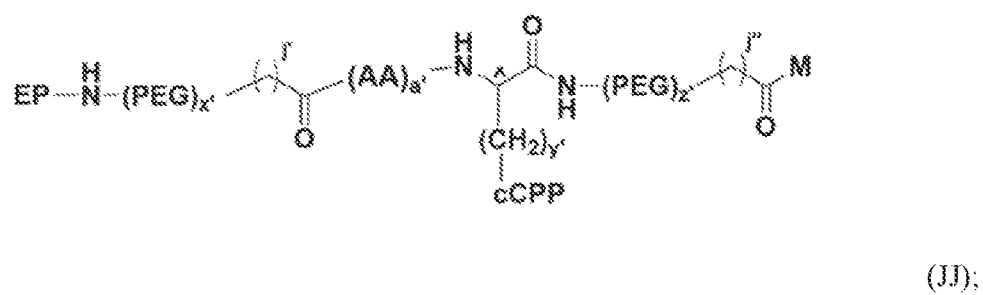


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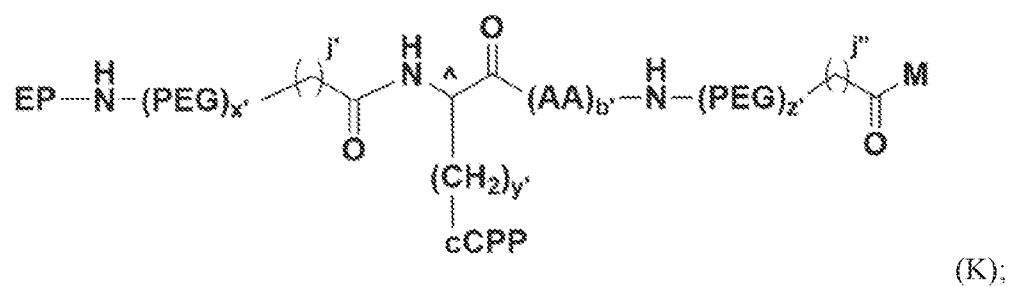
Formula (J):



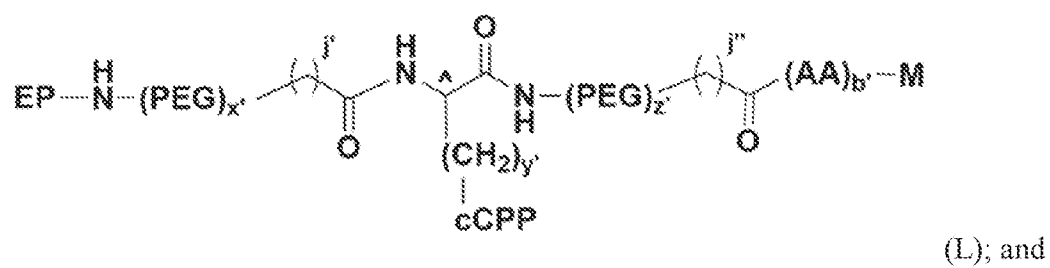
Formula (JJ):



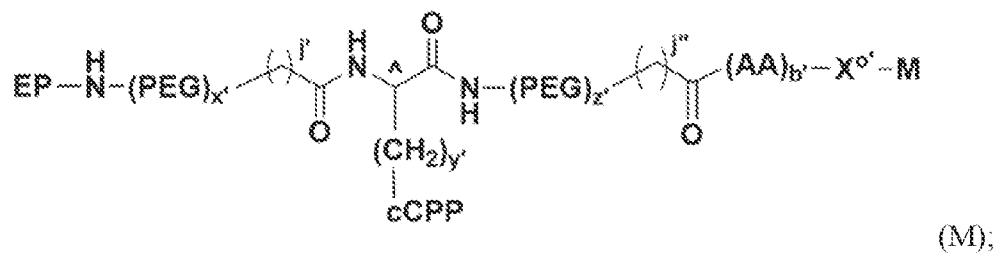
5 Formula (K):



Formula (L):



Formula (M):



wherein:

x' is an integer from 0 to 12;

5 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

10 $\text{X}^{o'}$ comprises a hydrophobic component;

$\text{K}^\#$ is D-lysine or L-lysine residue;

$(\text{AA})_{a'}$, $(\text{AA})_{b'}$, and $(\text{AA})_{b''}$ are each an amino acid (AA) component comprising at least one amino acid residue;

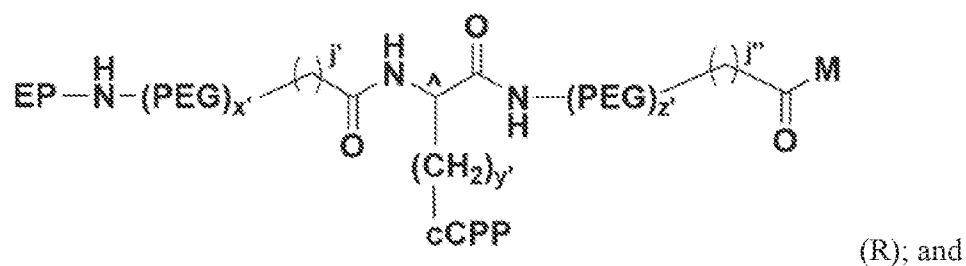
a' is an integer from 0 to 12;

15 b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

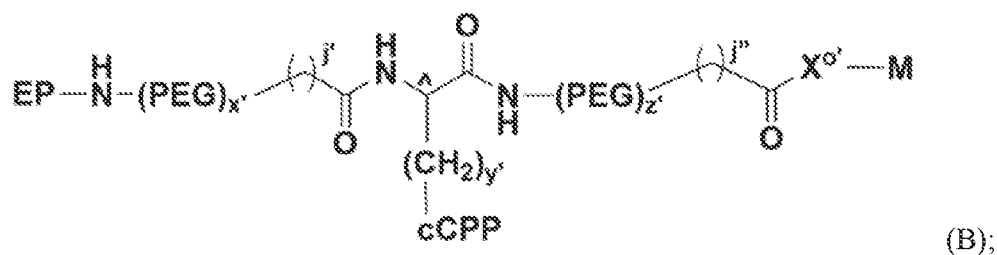
28. The EEV of claim 1, comprising a structure selected from:

Formula (R):



20

Formula (B):



wherein:

x' is an integer from 0 to 12;

5 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

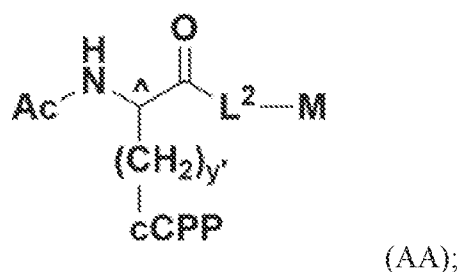
j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0; and

$X^{o'}$ comprises a hydrophobic component.

10 29. The EEV of claims 1-28, wherein EP comprises all D-amino acids; and cCPP comprises D-amino acids, achiral amino acids and AAsc, wherein AAsc is an amino acid side chain and AAsc conjugates the cCPP to the linker.

30. An EEV comprising the structure of Formula (AA):

15 Formula (AA):



wherein:

Ac is an acetyl group;

cCPP is a cyclic cell penetrating peptide;

20 L^2 is a linker arm;

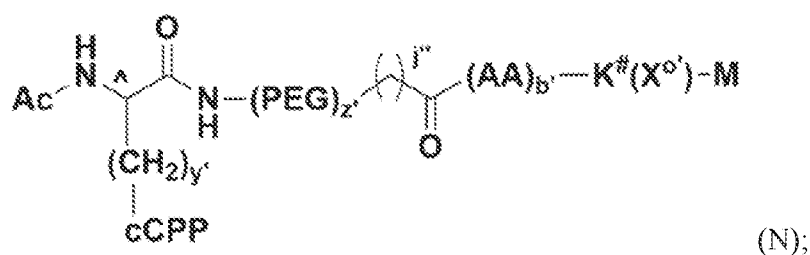
^ indicates L- or D-stereochemistry

y' is an integer from 1 to 5; and

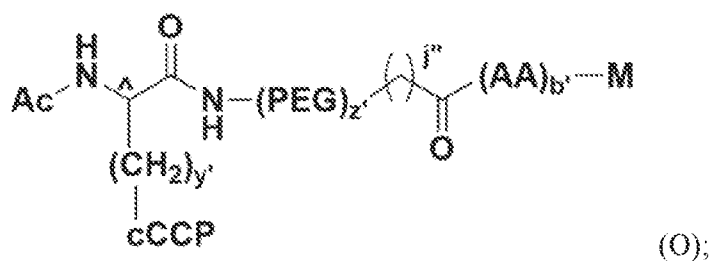
M is a reactive handle.

31. The EEV of claim 30, comprising a structure selected from:

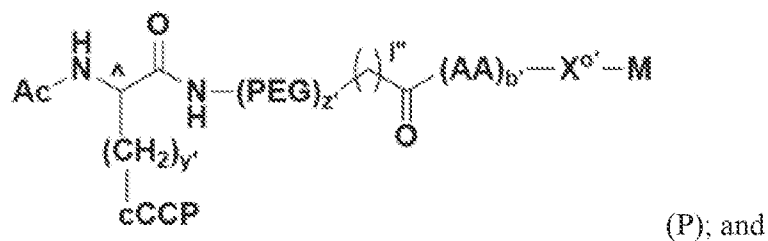
Formula (N):



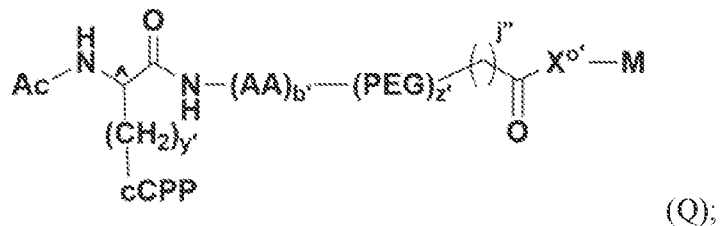
Formula (O):



Formula (P):



10 Formula (Q):



wherein:

z' is an integer from 0 to 12;

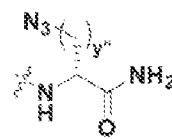
j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

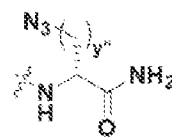
X^{or} is a hydrophobic component;

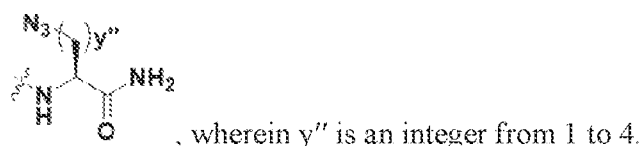
$K^{\#}$ is D-lysine or L-lysine residue;

(AA) b' is an amino acid (AA) component comprising at least one amino acid residue; and

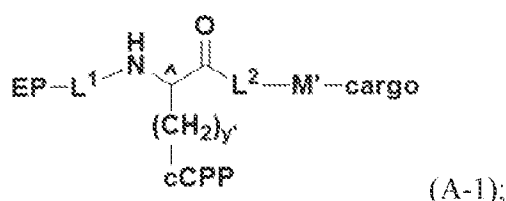
b' is an integer from 0 to 12.



32. The EEV of any of claims 1 to 31, wherein M comprises -OH, , or



33. A compound comprising an EEV-cargo conjugate having the structure of Formula (A-1):



wherein:

EP is a linear exocyclic peptide;

cCPP is a cyclic cell penetrating peptide;

L^1 and L^2 , are, independently, a linker arm;

$\wedge$ indicates D- or L-stereochemistry;

y' is an integer from 1 to 5;

M' is a bonding group.

34. The compound of claim 33, wherein L^1 comprises a polyethylene glycol component (PEG) x' , an amino acid component (AA) a' , or a combination thereof, wherein a' is an integer from 0-12 and x' is an integer from 0-12.

35. The compound of claim 33 or 34, wherein L^1 comprises (PEG) x' and x' is an integer from 1 to 12.

36. The compound of claim 34, wherein x' is an integer from 0, 2, 4, 8, or 12.
37. The compound of any of claims 34-36, wherein x' is 0 or 2.
- 5 38. The compound of any of claims 34-37, wherein (AA) a' comprises one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof.
39. The compound of claim 34, wherein (AA) a' comprises at least one charged amino acid.
- 10 40. The compound of claim 38 or 39, wherein a' is an integer from 1-12.
41. The compound of claim 38 or 39, wherein a' is an integer from 1-8.
- 15 42. The compound of claim 38 or 39, wherein a' is an integer from 2-5.
43. The compound of any of claims 33-42, wherein L^2 comprises one or more polyethylene glycol components (PEG) z' , (PEG) z'' , one or more amino acid components (AA) b' , (AA) b'' , one or more hydrophobic components ($X^{o'}$), ($X^{o''}$), or a combination thereof, wherein b' and b'' are,
- 20 independently, an integer from 0-12 and z' and z'' are, independently, an integer from 0-12.
44. The compound of claim 43, wherein L^2 comprises (PEG) z' , (PEG) z'' , or a combination thereof and z' and z'' are, independently, an integer from 1 to 12.
- 25 45. The compound of claim 43, wherein L^2 comprises (PEG) z' , (PEG) z'' , or a combination thereof and z' and z'' are, independently, 2, 4, 8, or 12.
46. The compound of claim 43, wherein L^2 comprises (PEG) z' , and z' is 2, 4, 8, or 12.
- 30 47. The compound of claim 43, wherein L^2 comprises (PEG) z' and (PEG) z'' and z' and z'' are, independently, 2, 4, 8, or 12.

48. The compound of any of claims 43-47, wherein (AA)b' and (AA)b'' comprise one or more amino acid residues selected from lysine (K), arginine (R), histidine (H), glycine (G), β -alanine (B), phenylalanine (F), proline (P), valine (V), or combinations thereof.

5

49. The compound of claim 48, wherein (AA)b' and (AA)b'' comprise at least one charged amino acid.

50. The compound of claim 48 or 49, wherein b' and b'' are, independently, an integer from 1-12.

10

51. The compound of claim 48 or 49, wherein b' and b'' are, independently, an integer from 1-8.

52. The compound of claim 48 or 49, wherein b' and b'' are, independently, an integer from 2-5.

15

53. The compound of any of claims 44-52, wherein L^2 comprises one or more hydrophobic components ($X^{o'}$), ($X^{o''}$).

20

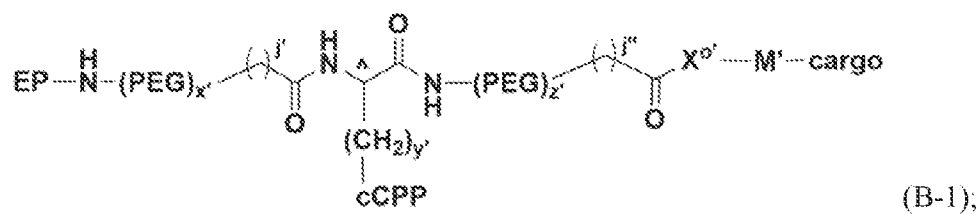
54. The compound of claim 53, wherein the hydrophobic components ($X^{o'}$), ($X^{o''}$) are the same.

55. The compound of claim 53, wherein the hydrophobic components ($X^{o'}$), ($X^{o''}$) are different.

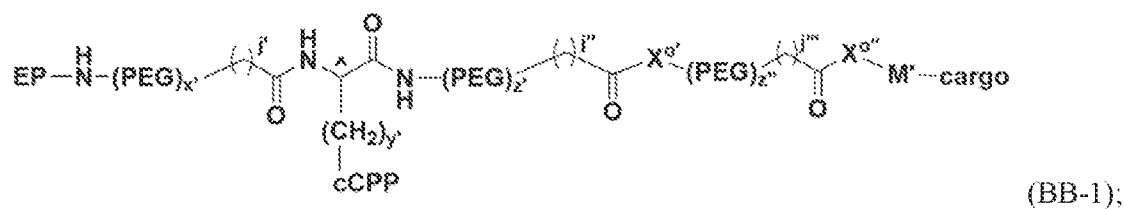
25

56. The compound of claim 33, comprising a structure selected from:

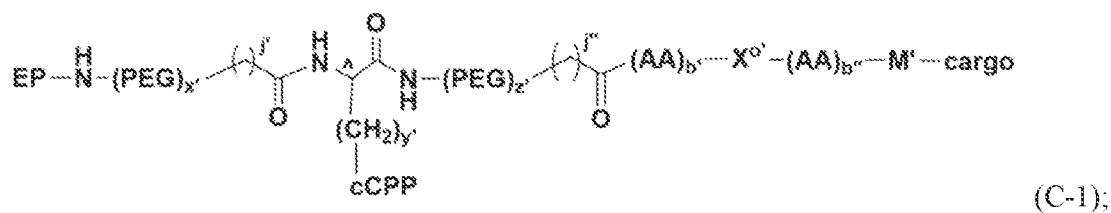
Formula (B-1):



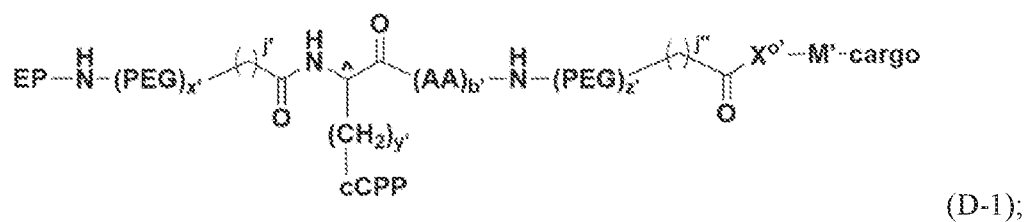
Formula (BB-1):



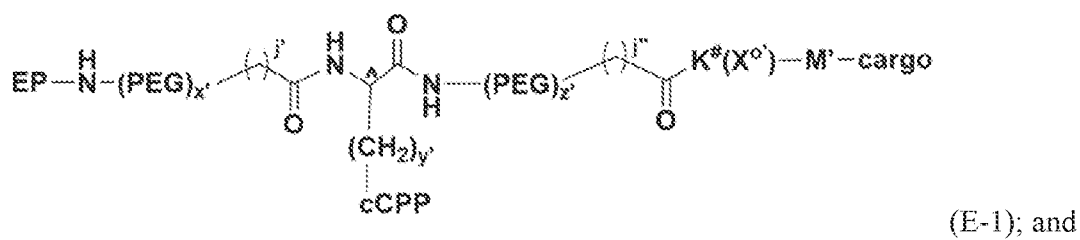
5 Formula (C-1):



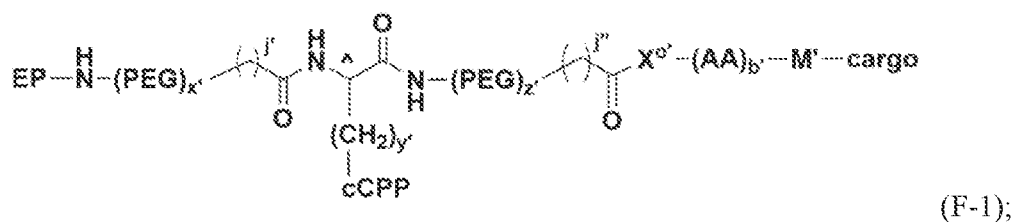
Formula (D-1):



Formula (E-1):



Formula (F-1):

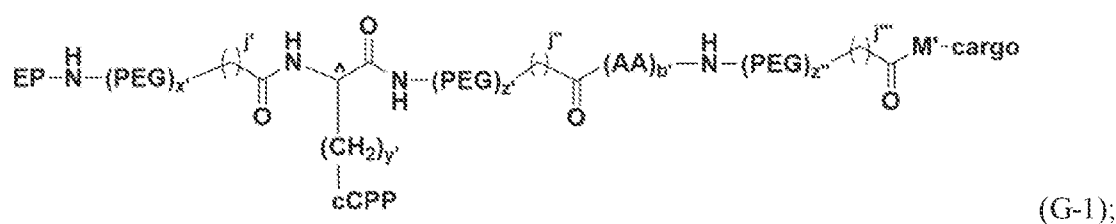


wherein:

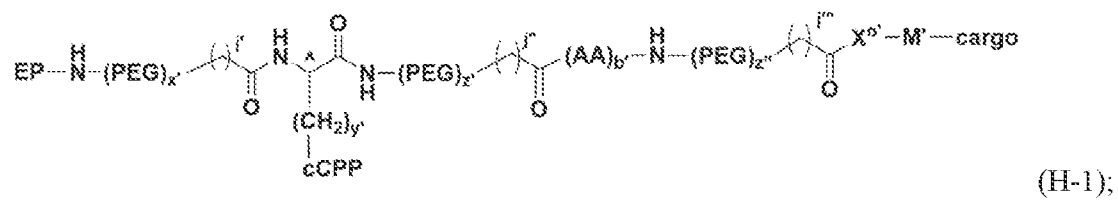
- x' is an integer from 0 to 12;
- 5 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;
- z' is an integer from 0 to 12;
- j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
- z'' is an integer from 0 to 12;
- j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;
- 10 $X^{o'}$ and $X^{o''}$ each, individually, comprise a hydrophobic component;
- $K^\#$ is D-lysine or L-lysine residue;
- $(\text{AA})_{b'}$ and $(\text{AA})_{b''}$ are each an amino acid (AA) component comprising at least one amino acid residue;
- b' is an integer from 0 to 12; and
- 15 b'' is an integer from 0 to 12.

57. The compound of claim 33, comprising a structure selected from:

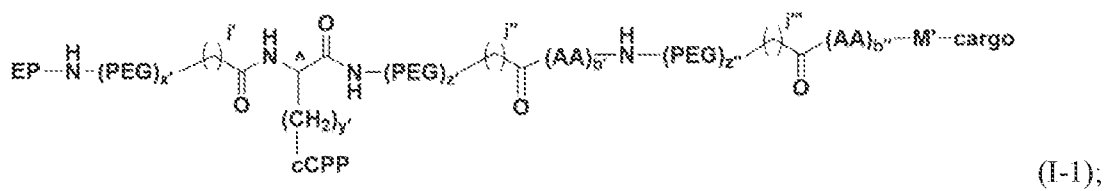
Formula (G-1):



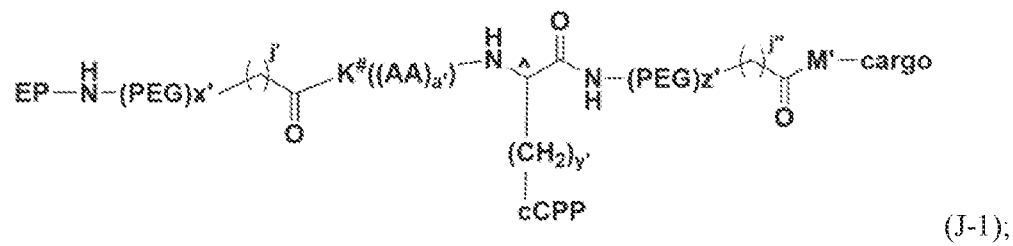
Formula (H-1):



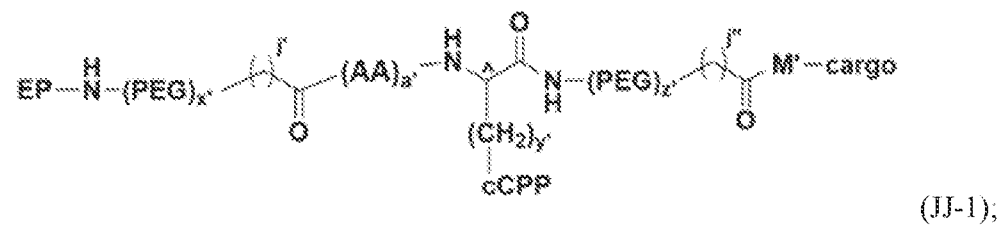
Formula (I-1):



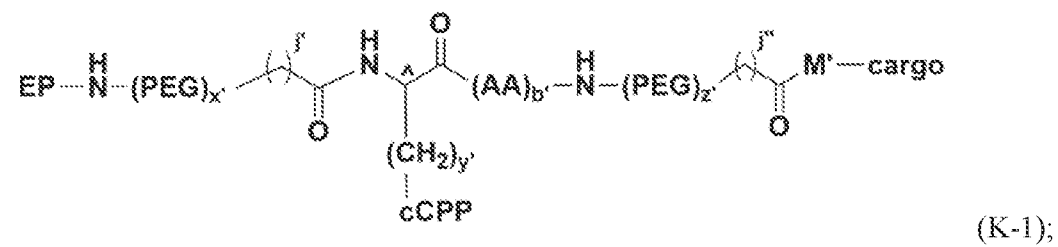
5 Formula (J-1):



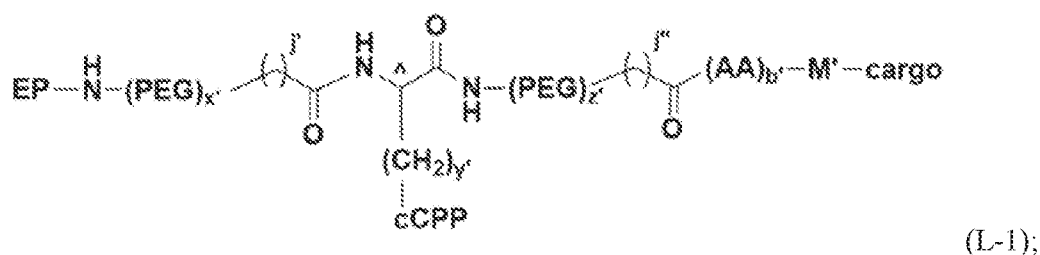
Formula (JJ-1):



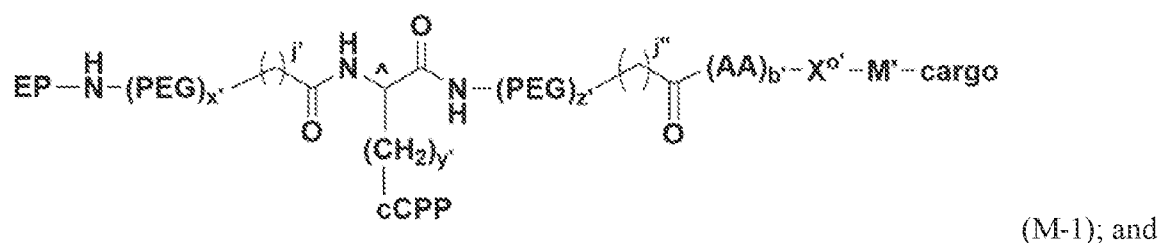
Formula (K-1):



Formula (L-1):



Formula (M-1):



5 wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

10 z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$\text{X}^{\alpha'}$ comprises a hydrophobic component;

$\text{K}^{\#}$ is D-lysine or L-lysine residue;

15 $(\text{AA})_{a'}$, $(\text{AA})_{b'}$, and $(\text{AA})_{b''}$ are each an amino acid (AA) component comprising at least one amino acid residue;

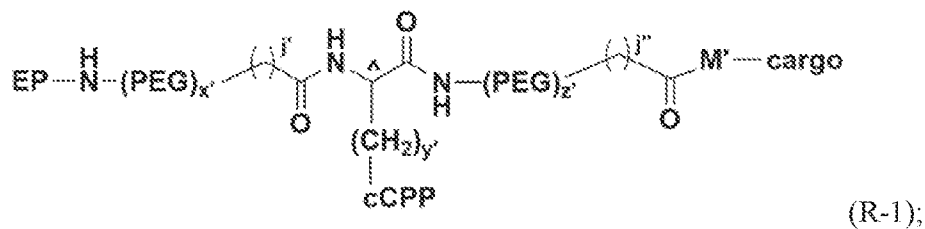
a' is an integer from 0 to 12;

b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

20 58. The compound of claim 33, comprising a structure selected from:

Formula (R-1):



x' is an integer from 0 to 12;

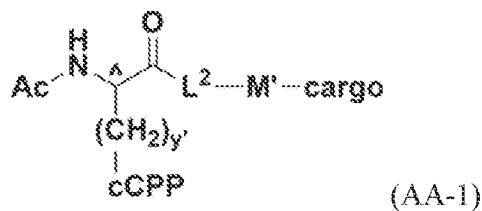
j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12; and

5 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0.

59. A compound comprising an EEV-cargo conjugate having the structure of Formula (AA-1):

Formula (AA-1):



wherein:

Ac is an acetyl group ;

cCPP is a cyclic cell penetrating peptide;

L^2 is a linker arm;

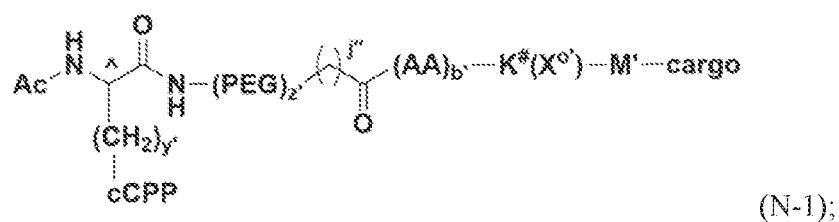
15 M' is a bonding group;

y' is an integer from 1 to 5; and

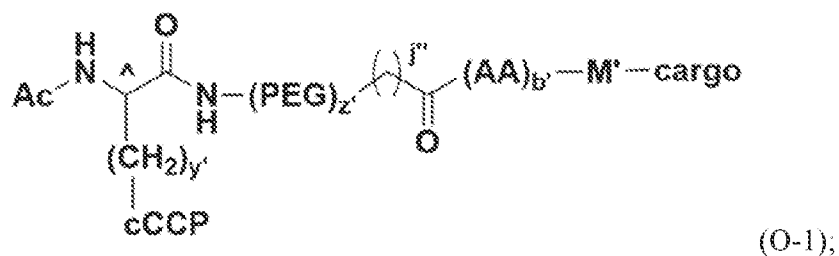
cargo is a peptide, oligonucleotide, a small molecule, or any combination thereof.

60. The compound of claim 59, comprising a structure selected from:

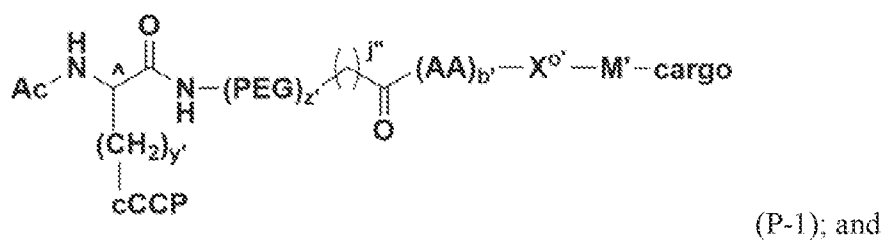
Formula (N-1):



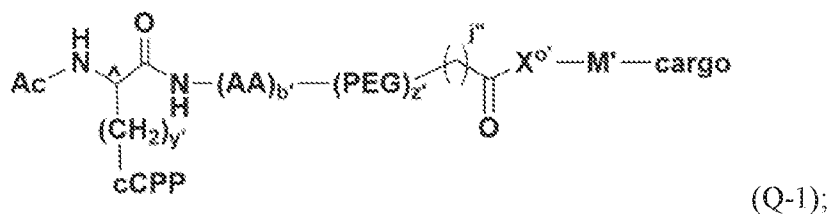
Formula (O-1):



5 Formula (P-1):



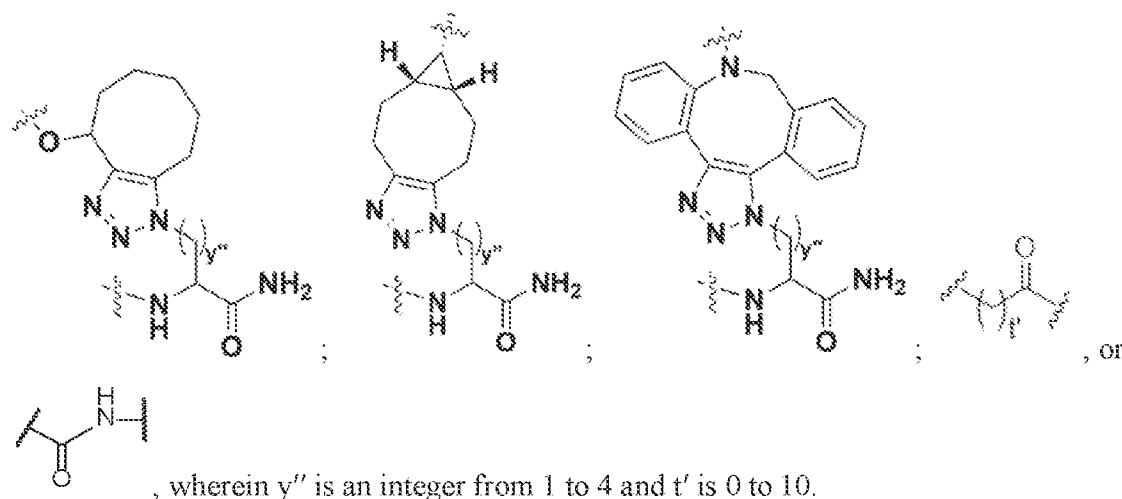
Formula (Q-1):



wherein:

- 10 z' is an integer from 0 to 12;
- j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
- $\text{X}^{\text{o}'}$ is a hydrophobic component;
- $\text{K}^{\#}$ is D-lysine or L-lysine residue;
- (AA) b' is an amino acid (AA) component comprising at least one amino acid residue; and
- 15 b' is an integer from 0 to 12.

61. The compound of claim 59, wherein the cargo is an oligonucleotide.
62. The compound of claim 61, wherein the oligonucleotide is an antisense oligonucleotide.
63. The compound of claim 61 or 62, wherein the oligonucleotide is a phosphorodiamidate morpholino oligonucleotide (PMO).
64. The compound of any of claims 61-63, wherein M' is covalently bound to the 5' end of the oligonucleotide, the 3' end of the oligonucleotide, or the backbone of the oligonucleotide.
65. The compound of any of claims 33 to 64, wherein M' comprises: -N(H), C(O), -O-,



66. The EEV or compound of the preceding claims, wherein the EP comprises from 2 to 10 amino acid residues, wherein at least 1 amino acid residue comprises a side chain comprising a guanidine group, a terminal amine, an imidazole, or a protonated form thereof.
67. The EEV or compound of claim 66, wherein the EP comprises from 2 to 8 amino acid residues.

68. The EEV or compound of claim 66, wherein the EP comprises from 2 to 6 amino acid residues.

69. The EEV or compound of any of claims 66 to 68, wherein the EP comprises 1, 2, 3, or 4 arginine residues.

70. The EEV or compound of any of claims 66 to 69, wherein the EP comprises 1, 2, 3, or 4 lysine residues.

71. The EEV or compound of any of claims 66 to 70, wherein the EP comprises 1 or 2 uncharged hydrophobic amino acid residues.

72. The EEV or compound of claim 71, wherein the uncharged hydrophobic amino acid residue is selected from valine, proline, b-alanine, glycine or a combination thereof.

73. The EEV or compound of claim 66, wherein the EP comprises a sequence selected from:

- (a) HBH, HBHBH, or HBRBH;
- (b) HHRBFBR;
- (c) KBK, KBKBK or KBR;
- (d) KGK or KGKK, KKGK;
- (e) KKKK, KKRK, KKKKR, KKKRK, KKKRKG, or KKRK;
- (f) KR, KRK, or KRKKK;
- (g) PGKKRKV;
- (h) PKGKRKV, PKK, PKKGRKV, PKKK, PKKKGKV, PKKKRGV, PKKKRKG, PKKRGV, PKKRKG, or PKKRKV;
- (i) RBHBH, RBHBR, RBR, RBRBR, or RBRRBR;
- (j) RFR;
- (k) RHR;
- (l) RK or RKKK;
- (m) RR, RF, RRR, RFRRF, or RRRRFFF;
- (n) RIRRI;

(o) KRKIL; and

(p) KFK, KRKF, or KKFRK;

wherein the amino acid residues in the EP can have D- or L-stereochemistry.

5 74. The EEV or compound of claim 66, wherein the EP has the structure: Ac-PKKKRKV-.

75. The EEV or compound of any of the preceding claims, wherein L^2 comprises one or more hydrophobic components ($X^{o'}$), ($X^{o''}$).

10 76. The EEV or compound of claim 75, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) comprise one or two aromatic rings.

77. The EEV or compound of claim 75, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) is a non-naturally occurring amino acid comprising one or more aromatic rings.

15 78. The EEV or compound of claim 75, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) comprise two aromatic rings.

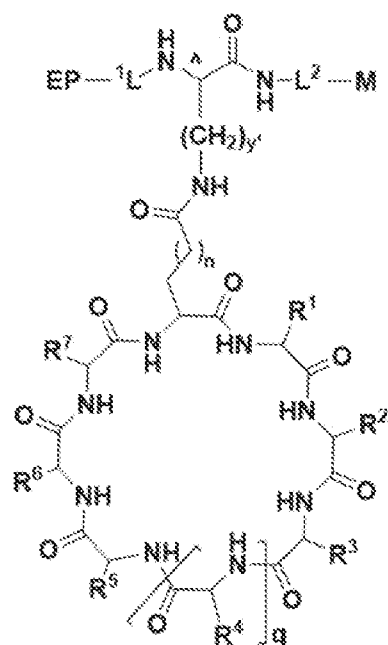
79. The EEV or compound of claim 75, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) is, independently, selected from Nal, dNal, or Bip.

80. The EEV or compound of claim 75, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) comprise an aliphatic hydrocarbon.

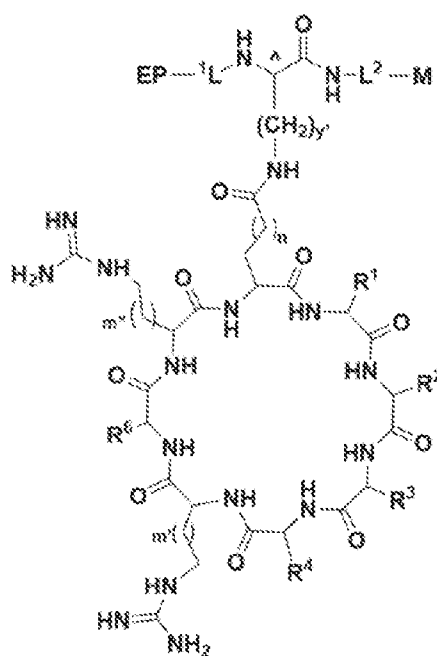
25 81. The EEV or compound of claim 80, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) comprises a C_4 - C_8 aliphatic hydrocarbon.

82. The EEV or compound of claim 81, wherein one or more hydrophobic components ($X^{o'}$), ($X^{o''}$) is a C_6 aliphatic hydrocarbon.

30 83. An EEV of claim 1 comprising the structure:



(A-2); or



(A-2a)

wherein,

R_1 , R_2 , and R_3 are each independently H or a side chain comprising an aryl or heteroaryl group;

at least two of R_1 , R_2 , and R_3 are a side chain of phenylalanine;

R_4 , R_5 , R_6 and R_7 , when present, are independently H or an amino acid side chain;

q is an integer from 1-4;

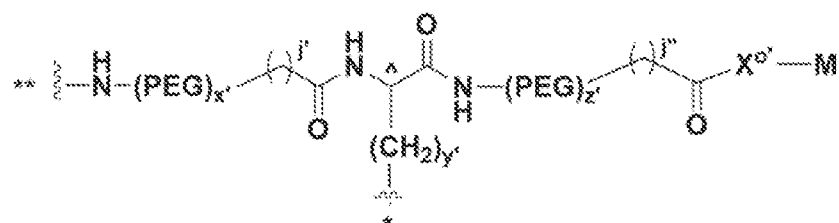
m' is an integer from 0 to 3;

m'' is an integer from 0 to 3; and

n is an integer from 1-4.

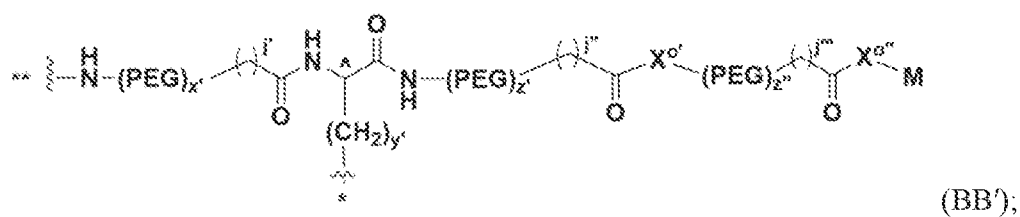
84. The EEV of claim 83, wherein the linker has a structure of Formula (B'), (BB'), (C'), (D'), (E') or (F'):

Formula (B'):

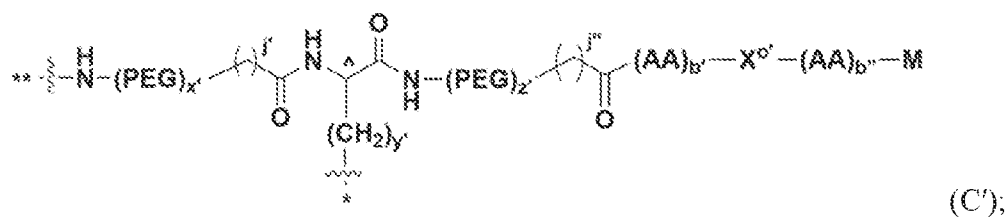


(B');

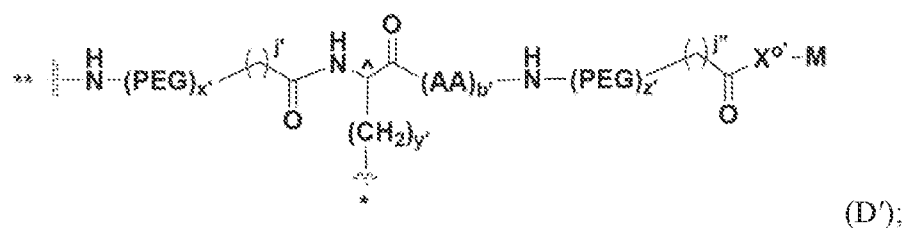
Formula (BB'):



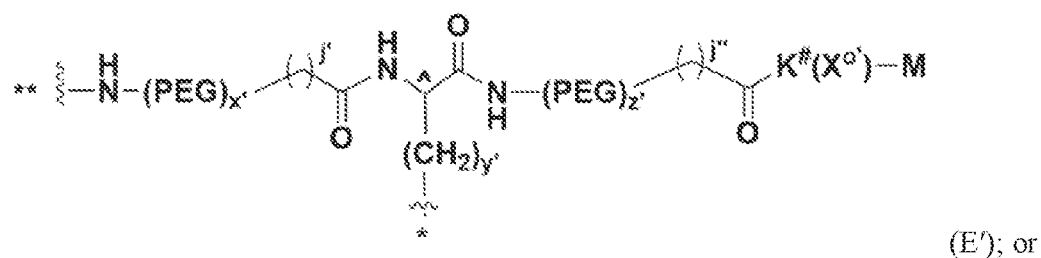
Formula (C'):



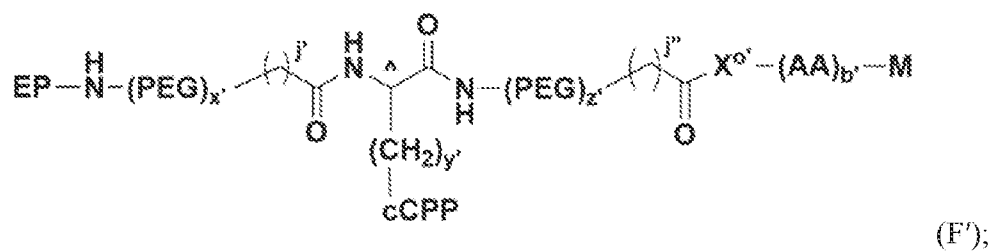
5 *Formula (D')*:



Formula (E'):



Formula (F'):



wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

5 z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ and $X^{o''}$ each, individually, comprise a hydrophobic component;

$K^\#$ is D-lysine or L-lysine residue;

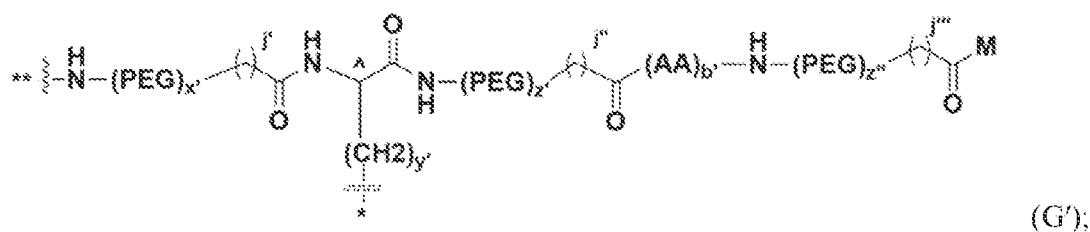
10 $(AA)b'$ and $(AA)b''$ are each an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

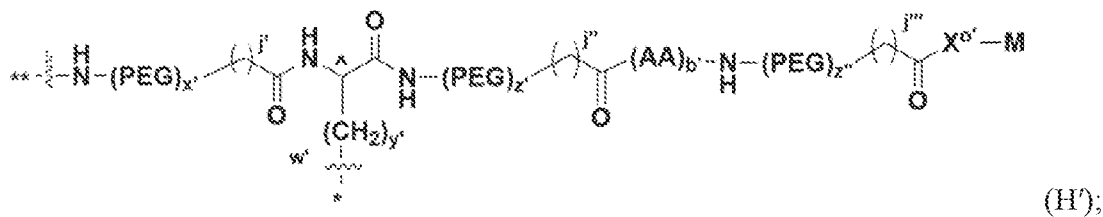
b'' is an integer from 0 to 12.

85. The EEV of claim 83, wherein the linker has a structure selected from Formula (G'), (H'),
15 (I'), (J'), (JJ'), (K'), (L'), or (M'):

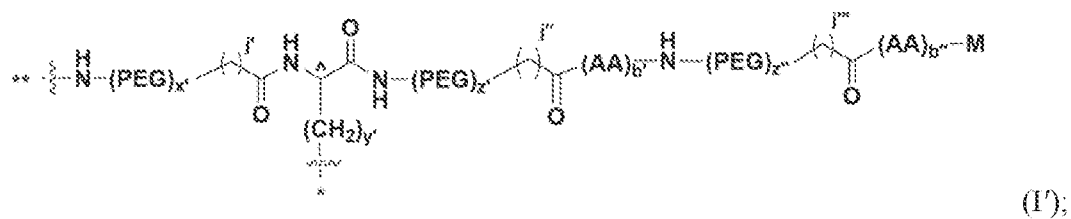
Formula (G'):



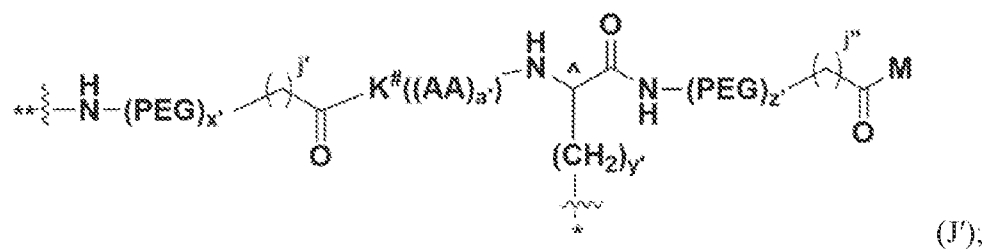
Formula (H'):



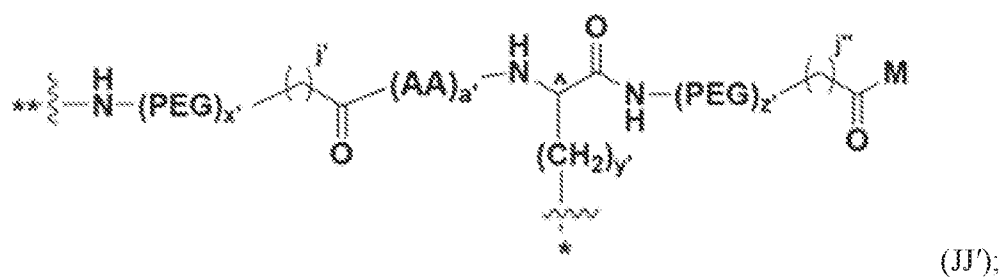
Formula (I):



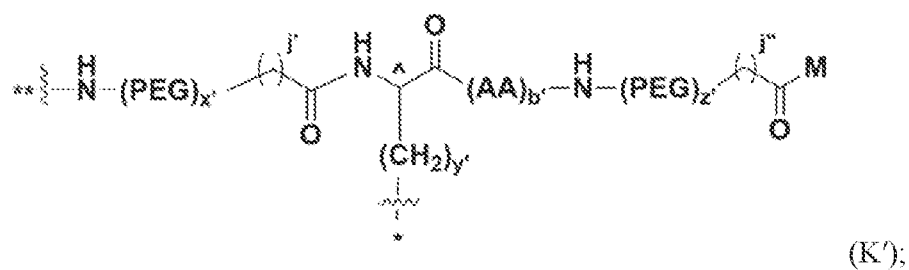
Formula (J):



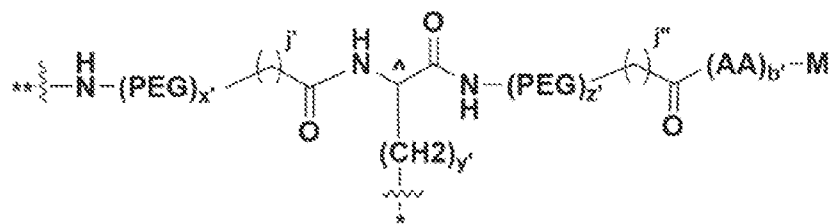
5 Formula (JJ):



Formula (K):

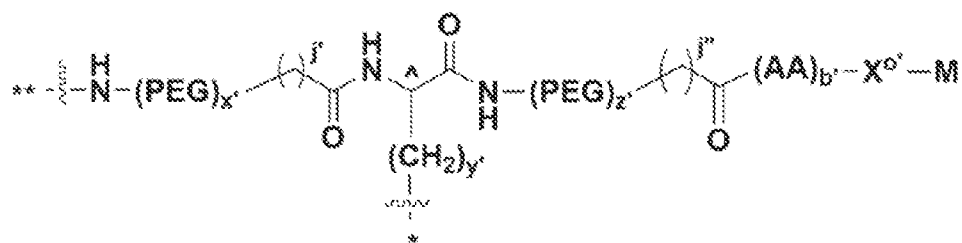


Formula (L'):



(L'); and

Formula (M'):



(M');

5 wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

10 z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ comprises a hydrophobic component;

$K^\#$ is D-lysine or L-lysine residue;

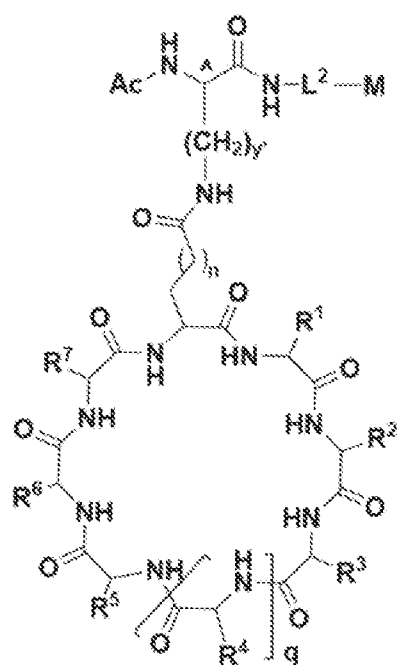
15 $(AA)a'$, $(AA)b'$, and $(AA)b''$ are each an amino acid (AA) component comprising at least one amino acid residue;

a' is an integer from 0 to 12;

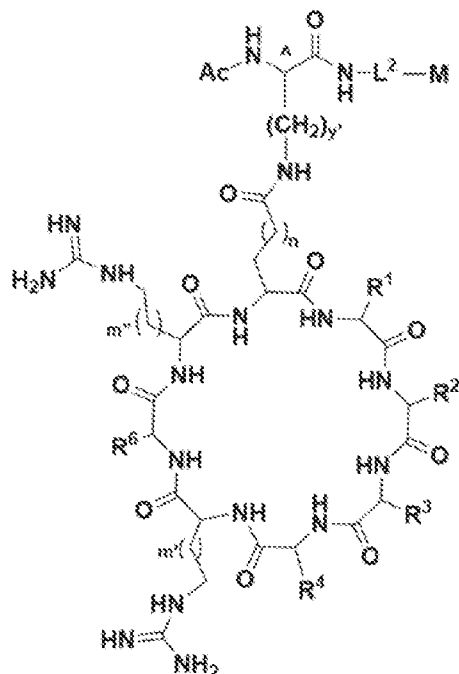
b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

20 86. An EEV of claim 30, comprising the structure:



(AA-2); or



(AA-2a);

wherein,

R_1 , R_2 , and R_3 are each independently H or a side chain comprising an aryl or heteroaryl group;

at least two of R_1 , R_2 , and R_3 are a side chain of phenylalanine;

R_4 , R_5 , R_6 and R_7 , when present, are independently H or an amino acid side chain;

q is 1, 2, 3 or 4;

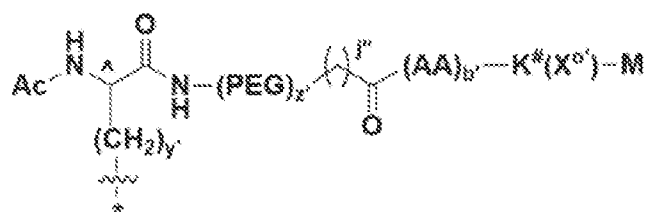
m' is an integer from 0 to 3;

m'' is an integer from 0 to 3; and

n is an integer from 1-4.

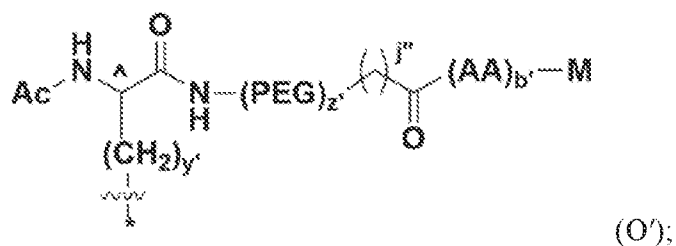
87. The EEV of claim 86, wherein the linker is selected from:

Formula (N'):

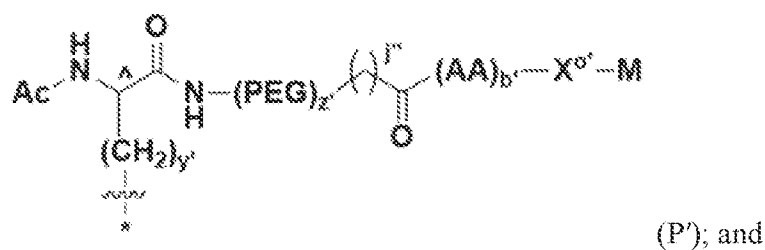


(N');

Formula (O'):



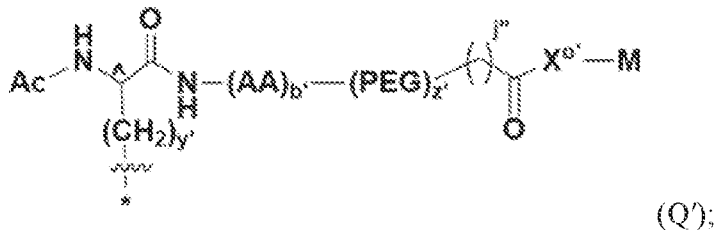
Formula (P'):



5

(P'); and

Formula (Q'):



wherein:

z' is an integer from 0 to 12;

10 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^{o'} is a hydrophobic component;

K[#] is D-lysine or L-lysine residue;

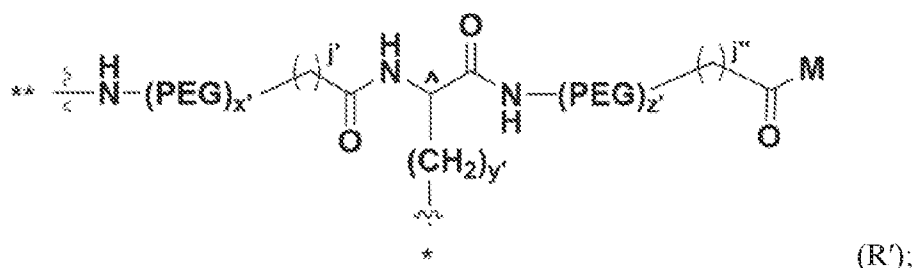
(AA)_{b'} is an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

15

88. The EEV of claim 83, wherein the linker comprises Formula (R'):

Formula (R'):



wherein:

x' is an integer from 0 to 12;

5 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12; and

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0.

89. The EEV of any of claims 83-88, wherein:

10 one of R₁, R₂, and R₃ is H; and

two of R₁, R₂, and R₃ are a side chain of phenylalanine.

90. The EEV of any of claims 83-88, wherein:

R₁, R₂, and R₃ are a side chain of phenylalanine.

15

91. The EEV of any of claims 83-88, wherein:

R₁, R₂, and R₃ are each independently a side chain comprising an aryl or heteroaryl group; and

at least two of R₁, R₂, and R₃ are a side chain of phenylalanine.

20

92. The EEV of any of claims 83-91, wherein R₄ and R₆ are H or an amino acid side chain of arginine, serine, histidine or citrulline.

93. The EEV of any of claims 83-92, wherein R₄ and R₆ are H or an amino acid side chain of arginine.

25

94. The EEV of any of claims 83-93, wherein R₄ and R₆ are H.

95. The EEV of any of claims 83-94, wherein R₅ and R₆ are arginine.

96. The EEV or compound of any of claims 1 to 88, wherein the cyclic cell penetrating peptide is selected from Ff-Nal-GrGrQ; FfFGRGRQ; FGFGGRGRQ; GfFGrGrQ; FGFGRRRQ; and FGFRRRRQ.

97. The EEV or compound of any of claims 1 to 88, wherein the cyclic cell penetrating peptide is FAFARARQ.

98. The EEV or compound of claims 1 to 88, wherein the cyclic cell penetrating peptide is Ff-Nal-GrGrQ.

99. The EEV or compound of claims 1 to 88, wherein the cyclic cell penetrating peptide is selected from FfFGRGRQ, FfFRRRRQ, FfFRrRrQ, fffrrrrQ or FfFRrRrQ.

100. The EEV or compound of claims 1 to 88, wherein the cyclic cell penetrating peptide is FG-Nal-GRGRQ.

101. The EEV or compound of claims 1 to 88, wherein the cyclic cell penetrating peptide is selected from FGFGGRGRQ, fGfGrGrQ, fGfGrGrQ, FGFGGRGRQ, FGFGGRQ, fGfrrrrQ, FGFRRRRQ, or FGFRRRRQ.

102. The EEV or compound of any of claims 1 to 88, wherein the cyclic cell penetrating peptide is FGWRGRQ.

103. The EEV or compound of any of claims 1-88, wherein the cyclic peptide is selected from FfFGRGRQ, FGFGGRGRQ, and FfFGRGRQ.

104. The EEV or compound of any of claims 1-88, wherein the cyclic peptide is RGRGRGRQ.

105. The EEV or compound of any of claims 1-88, wherein the cyclic peptide is FFFGRGRQ.

106. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is FGFRRRRQ.

5

107. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is FGFRGRGQ.

108. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is

10 FRGRGRGQ.

109. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is FRFGGRGRQ.

110. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is FRFRFRFQ.

15

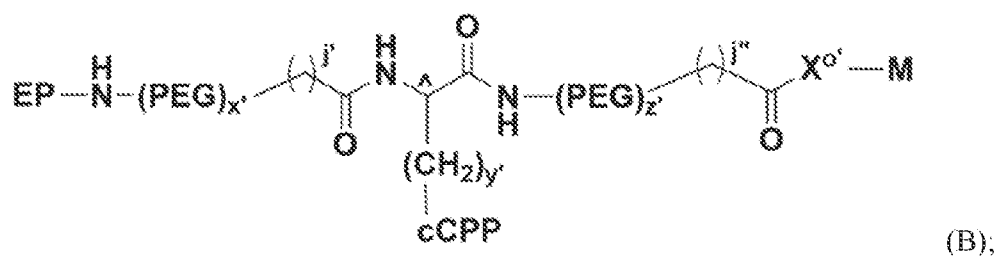
111. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is FGFLRLRQ.

112. The EEV on compound of any of claims 1-88, wherein the cyclic peptide is FGFVRVRQ.

20

113. The EEV of claim 26, comprising Formula (B):

Formula (B):



25

wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;
 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
 $X^{o'}$ comprises a hydrophobic component;
 b' is an integer from 0 to 12; and
 b'' is an integer from 0 to 12.

114. The EEV of claim 113, wherein $X^{o'}$ is Nal.

115. The EEV of claim 114, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFRRRGQ])-PEG ₁₂ -Nal-OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -Nal-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -Nal-OH
Ac-PKKKRKV-K(cyclo[fFRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FAFARARQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGFGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FGWRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-OH
Ac-PKKKRKV-K(cyclo[FG-Nal-GRGRQ])-PEG ₁₂ -Nal-OH; and
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrQ])-PEG ₁₂ -Nal-OH.

116. The EEV of claim 114, selected from:

Ac-RBRRBR-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₁₂ -Nal-OH
--

Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRRRRQ])-PEG ₁₂ -Nal-OH
Ac-RBRRBR-PEG ₂ -K(cyclo[FfFRrRrQ])-PEG ₁₂ -Nal-OH; and
Ac-rbrrbr-PEG ₂ -K(cyclo[fGfGrGrQ])-PEG ₁₂ -Nal-OH.

117. The EEV of claim 113, comprising:

Ac-rbrrbr-PEG₂-K(cyclo[fGfGrGrQ])-PEG₁₂-Nal-OH.

5 118. The EEV of claim 113, comprising:

Ac-pkkkrkv-PEG₂-k(cyclo[fGfrrrrQ])-PEG₁₂-Nal-OH.

119. The EEV of claim 113, wherein X^{o'} is dNal.

10 120. The EEV of claim 119, selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfrrrrQ])-PEG ₁₂ -dNal-OH
Ac-pkkkrkv-PEG ₂ -k(cyclo[ffrrrrQ])-PEG ₁₂ -dNal-OH
Ac-pkkkrkv-PEG ₂ -k(cyclo[FGFRRRRQ])-PEG ₁₂ -dNal-OH; and
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -dNal-OH.

121. The EEV of claim 119, comprising:

Ac-PKKKRKV-PEG ₂ -K(cyclo[RGRGRGRQ])-PEG ₁₂ -dNal-OH .
--

122. The EEV of claim 113, wherein X^{o'} is Bip.

15

123. The EEV of claim 122, comprising:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-Bip-OH.

124. The EEV of claim 113, wherein X^{o'} is a C₄-C₈ an aliphatic hydrocarbon.

20

125. The EEV of claim 124, comprising:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-C₆-OH.

126. The EEV of claim 113, wherein X^{o'} is a hydrophobic amino acid.

127. The EEV of claim 126, wherein the hydrophobic amino acid is selected from phenylalanine, tryptophan, tyrosine, valine, isoleucine, leucine, and histidine or a combination thereof.

128. The EEV of claim 127, wherein:

(a) z' is 12 and the EEV is selected from:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-F-OH;

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-W-OH;

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-Y-OH;

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-V-OH;

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-I-OH;

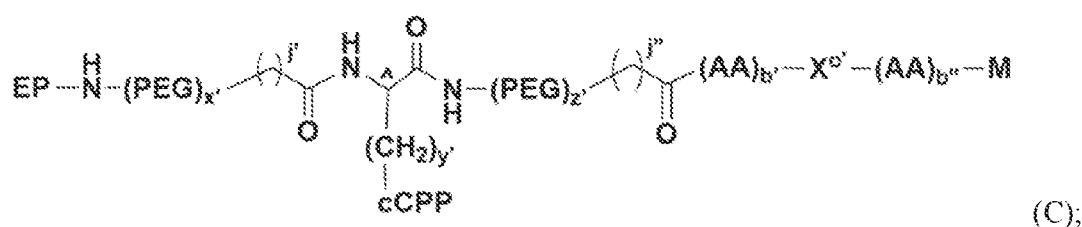
Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-L-OH;

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-H-OH; or

(a) z' is 0 and the EEV is Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-G-OH.

129. The EEV of claim 26, comprising Formula (C):

Formula (C):



wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^{o'} comprises a hydrophobic component;

(AA)b' and (AA)b'' each comprise an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

5

130. The EEV of claim 129, wherein b' is an integer from 2 to 5 and b'' is an integer from 1 to 8.

131. The EEV of claim 129, wherein b' is an integer from 1-8.

10

132. The EEV of claim 129, wherein b'' is an integer from 1-8.

133. The EEV of any one of claims 129, wherein b' is 2-5 and b'' is 2-5.

15

134. The EEV of claim 129, wherein x' is 2, z' is 12, b' is 2 and b'' is 2.

135. The EEV of any one of claims 130-133, wherein x' is 2.

136. The EEV of any one of claims 130-133, wherein z' is 12.

20

137. The EEV of any of claims 130-136, wherein the amino acid of (AA)b' comprises arginine, histidine, lysine, or a combination thereof.

138. The EEV of any one of claims 130 to 137, wherein the amino acids are D amino acids.

25

139. The EEV of any one of claims 130 to 137, wherein the amino acids are L amino acids.

140. The EEV of any one of claims 130 to 137, wherein X' is Nal, dNal, or Bip.

30

141. The EEV of claim 130 wherein X is Bip.

142. The EEV of claim 141, wherein the (AA)_{b'} comprises charged amino acids comprising histidine, asparagine, arginine, or combinations thereof.

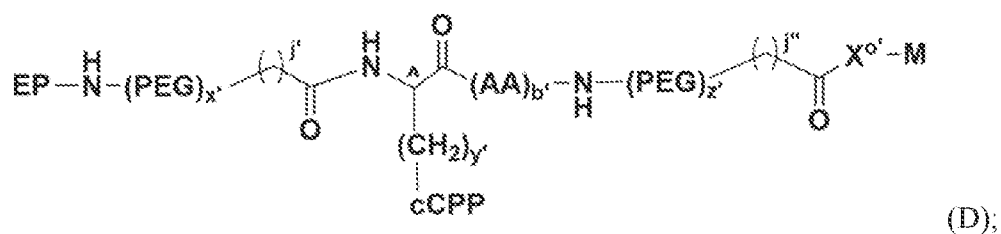
143. The EEV of claim 141, comprising:

Ac-PKKKRRKG-PEG2-K(cyclo[FGFGRGRQ])-PEG12-RB-Bip-RB-OH.

5

144. The EEV of claim 26, comprising Formula (D):

Formula (D):



10

wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

15

X^{o'} comprises a hydrophobic component;

(AA)_{b'} comprises an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

20

145. The EEV of claim 144, wherein b' is an integer from 2 to 5.

146. The EEV of claim 144 or 145, wherein (AA)_{b'} comprises charged amino acids comprising arginine, histidine, lysine, or combinations thereof.

25

147. The EEV of any one of claims 144-146, wherein the amino acids comprising (AA)_{b'} are D amino acids.

148. The EEV of any one of claims 144-147, wherein the amino acids comprising (AA)b' are L amino acids.

5 149. The EEV of any one of claims 144-148, wherein X^{o'} is Nal, dNal or Bip.

150. The EEV of claim 149, wherein X^{o'} is Nal.

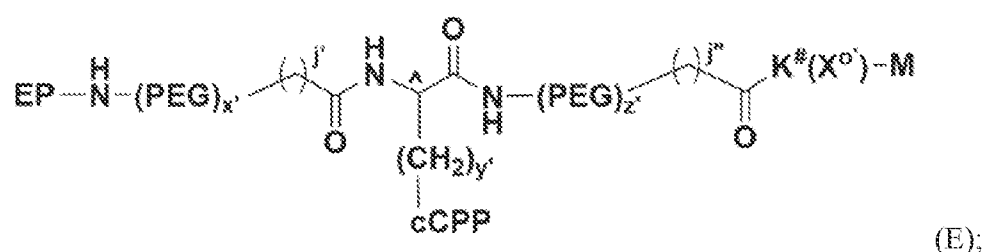
151. The EEV of claim 150, wherein (AA)b' comprises one or more R, K, or a combination thereof.

152. The EEV of claim 150, selected from:

Ac-KKK-PEG ₂ -K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-OH
Ac-KKK-K(cyclo[FGFGRGRQ])-RK-PEG ₁₂ -Nal-OH; and
Ac-K(cyclo[FGFGRGRQ])-KKKRK-PEG ₁₂ -Nal-OH.

153. The EEV of claim 26, comprising Formula (E):

15 *Formula (E):*



wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

20 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^{o'} comprises a hydrophobic component; and

K[#] is D-lysine or L-lysine residue.

154. The EEV of claim 153, wherein $X^{o'}$ comprises Nal, dNal, or Bip.

155. The EEV of claim 154, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Nal)-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Bip)-OH; and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkrk(Ac-nal)-OH.

5 156. The EEV of claim 153, wherein $X^{o'}$ is a hydrophobic amino acid.

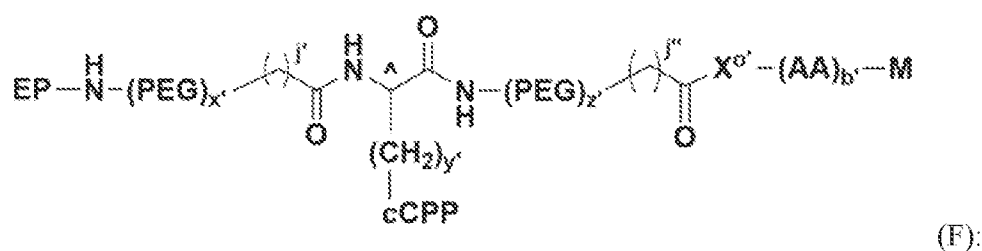
157. The EEV of claim 156, wherein the hydrophobic amino acid is tryptophan, tyrosine, isoleucine, leucine, histidine, phenylalanine, or a combination thereof.

10 158. The EEV of claim 156, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-W)-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-Y)-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-I)-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-L)-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-H)-OH; and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Ac-F)-OH.

159. The EEV of claim 26, comprising Formula (F):

Formula (F):



15 wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$X^{o'}$ comprises a hydrophobic component;

$(AA)b'$ comprises an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

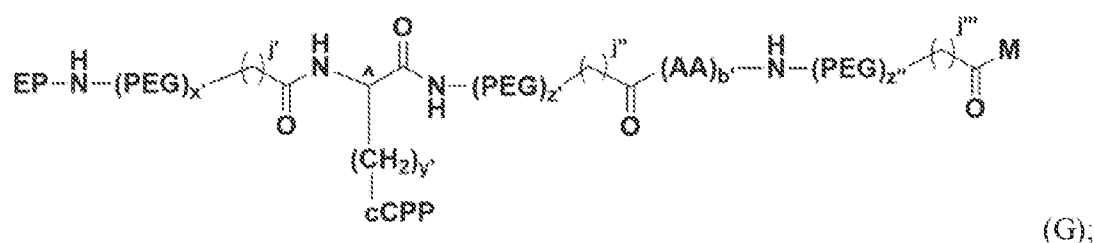
160. The EEV of claim 159, wherein $X^{o'}$ comprises Bip.

161. The EEV of claim 160, selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRBRB-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-RBRB-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BHRHR-OH; and
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -Bip-BRHRH-OH.

162. The EEV of claim 27, comprising Formula (G):

Formula (G):



wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

(AA) b' comprises an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

5 163. The EEV of claim 162, wherein x' is 2.

164. The EEV of claim 162 or 163, wherein z' is 2, 4, or 8.

165. The EEV of any of claims 162-164, wherein z'' is 2, 4 or 8.

10

166. The EEV of claim 162, wherein z' is 2 and z'' is 2.

167. The EEV of claim 162, comprising:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RFQILY-PEG ₂ -OH.
--

15 168. The EEV of claim 162, wherein z' is 2.

169. The EEV of claim 162, wherein z'' is 4.

170. The EEV of claim 162, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -R-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -F-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RRF-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RFR-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RGRRG-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RFRRF-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RBRRBR-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -KKKRK-PEG ₄ -OH; and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH.

20

171. The EEV of claim 162, comprising:



172. The EEV of claim 162, wherein z' is 4 and z'' is 4.

5

173. The EEV of claim 162, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₄ -RF-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -RF-PEG ₄ -OH; and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₄ -RF-PEG ₄ -OH.

174. The EEV of claim 162, wherein z' is 8 and z'' is 4.

10 175. The EEV of claim 162, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH; and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH.

176. The EEV of claim 162, selected from:

Ac-KK RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH;
Ac-KRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH;
Ac-RK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH; or
Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₈ -RF-PEG ₄ -OH.

177. The EEV of claim 162, selected from:

Ac-RF-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH; or
Ac-RF-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH.

15

178. The EEV of claim 162, selected from:

Ac-KKFRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH; or
Ac-KKFRK-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -OH.

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ comprises a hydrophobic component;

(AA) b' and (AA) b'' are each an amino acid (AA) component comprising at least one amino acid residue;

b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

183. The EEV of claim 182, wherein x' is 2.

184. The EEV of claim 182 or 183, wherein z' is 2, 4, or 8.

185. The EEV of any one of claims 182-184, wherein z'' is 2, 4, or 8.

186. The EEV of claim 182, wherein z' is 2, z'' is 2, and $X^{o'}$ is Bip.

187. The EEV of claim 186, comprising:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₂ -Bip-OH.
--

188. The EEV of claim 186, wherein z' is 2, z'' is 4 and $X^{o'}$ is Bip.

189. The EEV of claim 186, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-OH
Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-OH; and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -Bip-OH

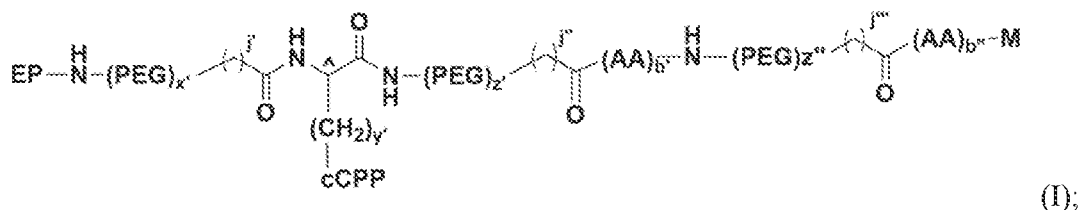
190. The EEV of claim 186, wherein z' is 2, z'' is 0, j''' is 0 and $X^{o'}$ is Bip.

191. The EEV of claim 186, comprising:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₈-R-Bip-OH.

192. The EEV of claim 27, comprising Formula (I):

Formula (I):



wherein:

- 5 x' is an integer from 0 to 12;
 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;
 z' is an integer from 0 to 12;
 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
 z'' is an integer from 0 to 12;
 10 j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;
 (AA)b' and (AA)b'' are each an amino acid (AA) component comprising at least one amino acid residue;
 b' is an integer from 0 to 12; and
 b'' is an integer from 0 to 12.

15

193. The EEV of claim 192, wherein x' is 2.

194. The EEV of claim 192 or 193 wherein z' is 2, 4, or 8.

20 195. The EEV of any one of claims 191-193, wherein z'' is 2, 4 or 8.

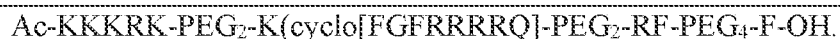
196. The EEV of claim 192, comprising:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH; or
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₂ -RF-PEG ₄ -R-OH.

197. The EEV of claim 192, comprising:

Ac-KKKRK-PEG ₂ -K(cyclo[FFFGRGRQ])-PEG ₂ -RF-PEG ₄ -F-OH.
--

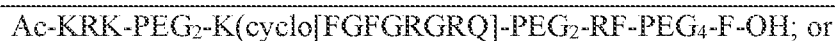
198. The EEV of claim 192, comprising:



199. The EEV of claim 192, comprising:

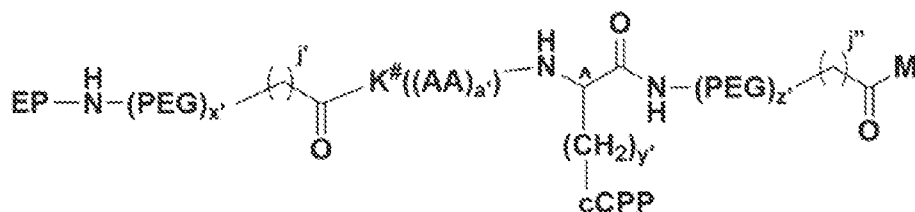


200. The EEV of claim 192, comprising:



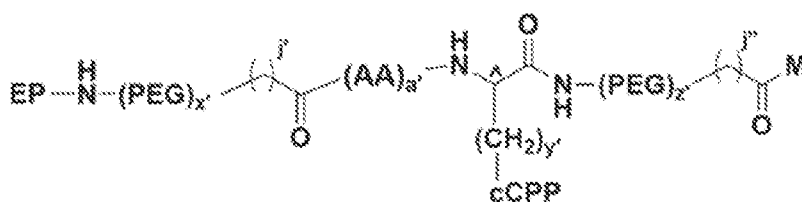
201. The EEV of claim 27, comprising Formula (J) or Formula (JJ):

Formula (J):



(J); or

Formula (JJ):



(JJ);

wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$K^\#$ is D-lysine or L-lysine residue;

(AA) a' comprises an amino acid (AA) component comprising at least one amino acid residue; and

5 a' is an integer from 0 to 12.

202. The EEV of claim 200, wherein x' is 2.

203. The EEV of claim 200 or 201, wherein z' is 12.

10

204. The EEV of claim 200, wherein x' is 2 and z' is 12.

205. The EEV of claim 200, selected from:

Ac-KKKRK-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂
Ac-KKKRK-PEG ₂ -K(RBRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂
Ac-RBRRBR-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂ ; and
Ac-KRK-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(N ₃)-NH ₂ .

15 206. The EEV of claim 200, selected from:

Ac-KRK-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH;
Ac-KRK-PEG ₂ -K(KRKIL-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH;
Ac-KRKIL-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH;
Ac-KRK-PEG ₂ -K(KRKF-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH; or
Ac-KRKF-PEG ₂ -K(KRK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH.

207. The EEV of claim 200, selected from:

Ac-KKKRK-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH;
Ac-KKKRK-PEG ₂ -K(RBRRBR-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH; or
Ac-RBRRBR-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH.

208. The EEV of claim 200, selected from:

Ac-KKKRK-PEG ₂ -K(RFRRF-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH; or
Ac-RFRRF-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH.

209. The EEV of claim 200, selected from:

Ac-KKKRK-PEG ₂ -K(RIRRI-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH; or
Ac-RIRRI-PEG ₂ -K(KKRKK-Ac)-K(cyclo[FGFGRGRQ])-PEG ₁₂ -OH.

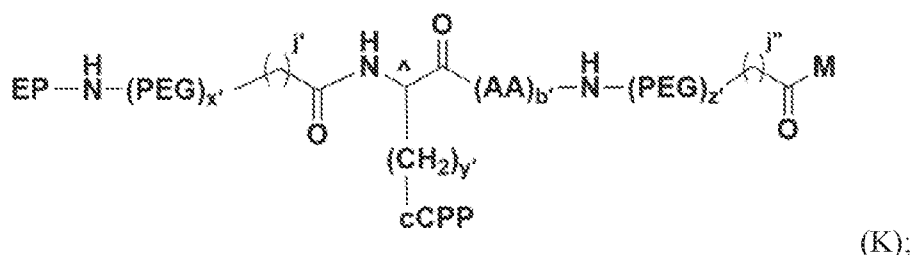
210. The EEV of claim 200, selected from:

Ac-PKKKRKV-PEG ₂ -FGF-K(cyclo[RGRGRGRQ])-PEG ₁₂ -OH.
--

5

211. The EEV of claim 27, comprising Formula (K):

Formula (K):



wherein:

- 10 (AA)_a' is an amino acid linker comprising at least one amino acid;
 x' is an integer from 0 to 12;
 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;
 z' is an integer from 0 to 12;
 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;
 15 (AA)_b' comprises an amino acid (AA) component comprising at least one amino acid residue; and
 b' is an integer from 0 to 12.

20

212. The EEV of claim 211, wherein b' is an integer from 1-8.

213. The EEV of claim 211-212, wherein the amino acid in (AA)_b' comprises G, F, B, R or combinations thereof.

214. The EEV of any of claims 211-213, wherein x' is 2.

215. The EEV of any of claims 211-214, wherein z' is 4.

5

216. The EEV of claim 211, wherein b' is 2, x' is 2 and z' is 4.

217. The EEV of claim 211, selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FfFGGRGRQ])-RF-PEG ₄ -OH; and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGGRGRQ])-RF-PEG ₄ -OH.

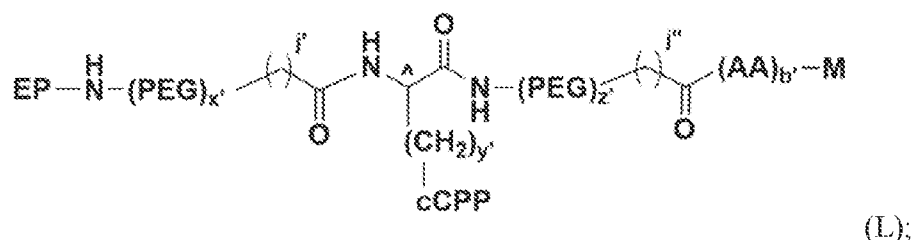
10 218. The EEV of claim 211, wherein a' is 3, x' is 2 and z' is 4.

219. The EEV of claim 211 selected from:

Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGGRGRQ])-GRF-PEG ₄ -OH; and
Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGGRGRQ])-BRF-PEG ₄ -OH.

220. The EEV of claim 27, comprising formula (L):

15 *Formula (L):*



wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

20 z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

(AA) b' comprises an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

221. The EEV of claim 220, wherein b' is an integer from 1-8.

5 222. The EEV of claim 220 or 221, wherein (AA)b' comprises a charged amino acid selected from G, E, R or a combination thereof.

223. The EEV of any one of claims 220-222, wherein x' is 2.

10 224. The EEV of any one of claims 220-223, wherein z' is 0, 2 or 8.

225. The EEV of claim 220, wherein x' is 2, z' is 0, j'' is 0 and b' is 3.

226. The EEV of claim 220, selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-GGE-OH; and
Ac-KKKRK-PEG ₂ -K(cyclo[FGFGRGRQ])-GGE-NH ₂ .

15

227. The EEV of claim 220, selected from:

Ac-KKKRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH;
Ac-KKKRK-PEG ₄ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH; or
Ac-KKKRK-PEG ₈ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH.

228. The EEV of claim 220, selected from:

Ac-KK RK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH;
Ac-KRK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH;
Ac-RK-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH; or
Ac-RF-PEG ₂ -K(cyclo[FfFGRGRQ])-PEG ₈ -RF-OH.

20 229. The EEV of claim 220, wherein x' is 2, z' is 0, j'' is 0, and b' is an integer from 2-8.

230. The EEV of claim 220, comprising:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-GGRRGRRG-OH.

231. The EEV of claim 220, wherein (AA)_{b'} comprises a charged amino acid selected from R and F.

5 232. The EEV of claim 227, wherein x' is 2, z' is 2, and b' is an integer from 8-12.

233. The EEV of claim 220, comprising:

Ac-PKKKRKV-PEG₂-K(cyclo[FfFGRGRQ])-PEG₂-RFQILYBRBRB-OH.

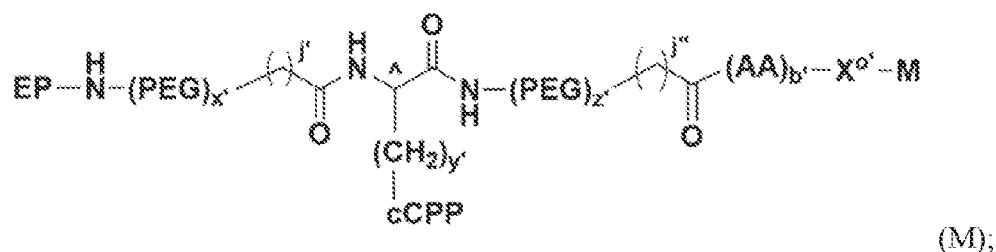
10 234. The EEV of claim 220, wherein x' is 2, z' is 8, and b' is 2.

235. The EEV of claim 220, comprising:

Ac-KKKRK-PEG₂-K(cyclo[FGFGRGRQ])-PEG₈-RF-OH.

15 236. The EEV of claim 27, comprising Formula (M):

Formula (M):



wherein:

x' is an integer from 0 to 12;

20 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^{o'} comprises a hydrophobic component;

(AA)_{b'} comprises an amino acid (AA) component comprising at least one amino acid
25 residue;

a' is an integer from 0 to 12;

b' is an integer from 0 to 12; and

b'' is an integer from 0 to 12.

237. The EEV of claim 236, wherein X^{o'} is Nal, dNal, or Bip.

5 238. The EEV of claim 236, wherein X^{o'} is Bip.

239. The EEV of claim 236, selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-OH
Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-OH

240. The EEV of claim 236, wherein the amino acid in of (AA)^{b'} comprises histidine,
10 asparagine, arginine, or combinations thereof.

241. The EEV of claim 236, selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RHRH-Bip-OH; and Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RBRB-Bip-OH.
--

242. The EEV of claim 236, selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BRBR-Bip-OH; and Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -BHBH-Bip-OH.
--

15

243. The EEV of claim 236, selected from:

Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HRHR-Bip-OH; and Ac-PKKKRKG-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -HBHB-Bip-OH.
--

244. The EEV of claim 236, wherein X^{o'} is Nal.

245. The EEV of claim 244, selected from:

Ac-KKK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-OH;
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -RK-Nal-OH;
Ac-KKK-PEG ₂ -K(cyclo[Ff-Nal-GrGrQ])-PEG ₁₂ -RK-Nal-OH;
Ac-KK-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKK-Nal-OH;
Ac-K-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKRK-Nal-OH; and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKRK-Nal-OH.

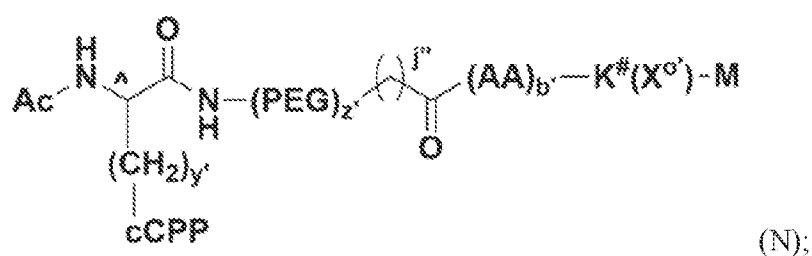
246. The EEV of claim 236, wherein X^{o'} is dNal.

5 247. The EEV of claim 246, selected from:

Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-OH;
Ac-KKK-K(cyclo[FGFGRGRQ])-PEG ₁₂ -rk-nal-OH;
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-OH; and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -kkkr-nal-OH.

248. The EEV of claim 31, comprising Formula (N):

Formula (N):



10 wherein:

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

X^{o'} is a hydrophobic component;

K[#] is D-lysine or L-lysine residue;

15 (AA)_{b'} is an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

249. The EEV of claim 248, wherein z' is 2, 4, 8, or 12.

250. The EEV of claim 248 or 249, wherein z' is 12.

5 251. The EEV of any of claims 248 to 250, wherein b' is an integer from 2-5.

252. The EEV of claim 248, selected from:

Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKKR-K(Ac-Nal)-OH;

Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKR-K(Ac-Nal)-OH; and

10 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KR-K(Ac-Nal)-OH.

253. The EEV of claim 248, selected from

Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKKR-K(Ac-dNal)-OH;

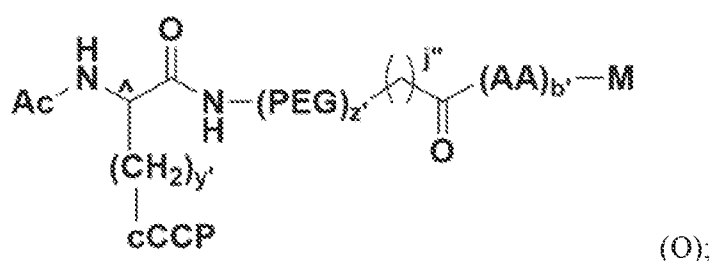
Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KKR-K(Ac-dNal)-OH; and

15 Ac-K(*cyclo*[FGFGRGRQ])-PEG₁₂-KR-K(Ac-dNal)-OH.

254. The EEV of claim 252 or 253, wherein (AA) b' comprises $N^{\alpha Me}$.

255. The EEV of claim 31, comprising Formula (O):

20 *Formula (O):*



wherein:

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

25 (AA) b' is an amino acid (AA) component comprising at least one amino acid residue; and
 b' is an integer from 0 to 12.

256. The EEV of claim 255, wherein z' is 2, 4, 8, or 12.

257. The EEV of claim 255 or 256, wherein z' is 12.

5 258. The EEV of any of claims 255-257, wherein b' is 2, 3, or 4.

259. The EEV of claim 255, selected from:

Ac-K(*cyclo*[FGFGGRGRQ])-PEG₁₂-KKKR-k(N₃)-NH₂

Ac-K(*cyclo*[FGFGGRGRQ])-PEG₁₂-KKR-k(N₃)-NH₂; and

10 Ac-K(*cyclo*[FGFGGRGRQ])-PEG₁₂-KR- k(N₃)-NH₂.

260. The EEV of claim 255, selected from:

Ac-K(*cyclo*[FGFGGRGRQ])-PEG₁₂-KKKR-OH

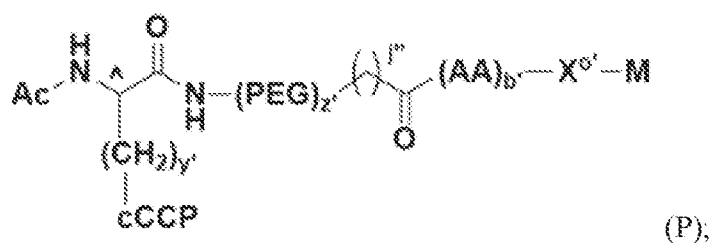
Ac-K(*cyclo*[FGFGGRGRQ])-PEG₁₂-KKR-OH; and

15 Ac-K(*cyclo*[FGFGGRGRQ])-PEG₁₂-KR-OH.

261. The EEV of claims 259 or 260, wherein (AA) b' comprises N^{Me} .

262. The EEV of claim 31, comprising Formula (P):

20 *Formula (P):*



wherein:

z' is an integer from 0 to 12;

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

25 $X^{o'}$ is a hydrophobic component;

(AA) b' is an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

263. The EEV of claim 262, wherein z' is 2, 4, 8, or 12.

264. The EEV of claim 262 or 263, wherein z' is 12.

5

265. The EEV of any of claims 262-264, wherein b' is 2-8.

266. The EEV of any of claims 262-265, wherein $X^{o'}$ is Nal or dNal.

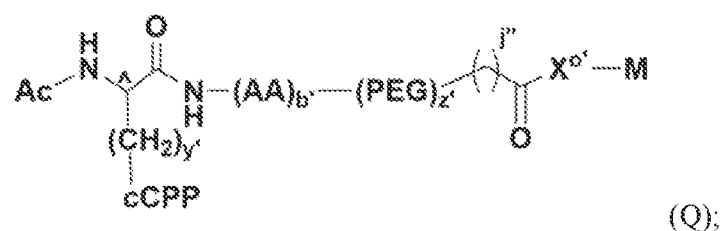
10 267. The EEV of claim 262, selected from:

Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKKR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-OH
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KR-Nal-OH; and
Ac-K(cyclo[FGFGRGRQ])-PEG ₁₂ -KKR-Nal-OH

268. The EEV of claim 267, wherein $(AA)_{b'}$ comprises N^{Me} .

269. The EEV of claim 31, comprising Formula (Q):

15 *Formula (Q):*



wherein:

z' is an integer from 0 to 12;

20 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

$X^{o'}$ is a hydrophobic component;

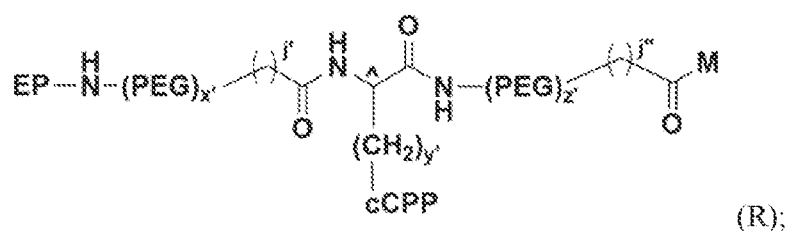
$(AA)_{b'}$ is an amino acid (AA) component comprising at least one amino acid residue; and

b' is an integer from 0 to 12.

270. The EEV of claim 265 comprising: Ac-K(cyclo[FGFGRGRQ])-KKKRK-PEG₁₂-Nal-OH.

5 271. The EEV of claim 28, comprising Formula (R):

Formula (R):



wherein:

x' is an integer from 0 to 12;

10 j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12; and

j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0.

272. The EEV claim 271, wherein x' is 2.

15

273. The EEV of claim 271 or 272, wherein z' is 2, 4, 8 or 12.

274. The EEV of any one of claims 271-273, wherein x' is 2 and z' is 12.

20 275. The EEV of claim 271, selected from:

Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -OH; and
Ac-pkkkrkv-PEG ₂ -k(cyclo[fGfGrGrQ])-PEG ₁₂ -K(N ₃)-NH ₂ .

276. The EEV of any of claims 1 to 32 or 66 to 271, conjugated to a cargo to form an EEV-cargo conjugate.

25 277. The EEV of claim 276, wherein cargo is a peptide, oligonucleotide, a small molecule, or

a combination thereof.

278. The EEV of claim 277, wherein cargo is an oligonucleotide.

5 279. The EEV of claim 278, wherein oligonucleotide is an antisense oligonucleotide.

280. The EEV of 278 or 279, wherein oligonucleotide is a phosphorodiamidate morpholino oligonucleotide (PMO).

10 281. The EEV of any of claims 278-280, wherein M is covalently bound to the 5' end of the oligonucleotide, the 3' end of the oligonucleotide, or the backbone of the oligonucleotide.

282. A pharmaceutical composition comprising the compound of claims 33-82 and a pharmaceutically acceptable carrier.

15

283. A method of treating a disease in a subject in need thereof comprising administering to the subject an effective amount of the pharmaceutical composition of claim 282.

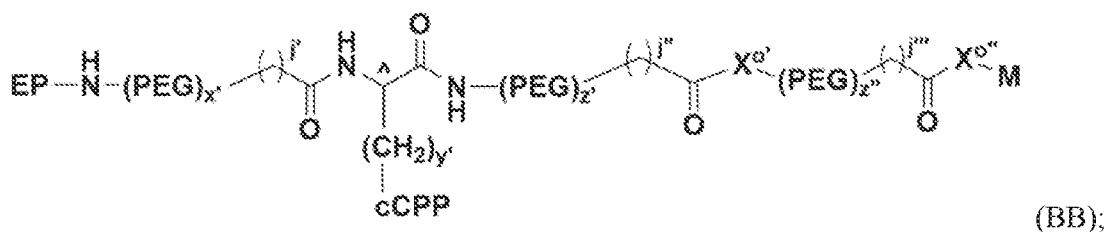
284. The method of claim 283, wherein administering comprises parenteral administration.

20

285. The method of claim 284, wherein parenteral administration is selected from: subcutaneous, intradermal, intravenous, intramuscular, intraperitoneal, intrasternal, intrathecal and intracerebroventricular administration.

25 286. The EEV of claim 26, comprising a structure selected from:

Formula (BB):



wherein:

x' is an integer from 0 to 12;

j' is 0, 1, or 2, wherein j' is 0 when x' is 0;

z' is an integer from 0 to 12;

5 j'' is 0, 1, or 2, wherein j'' is 0 when z' is 0;

z'' is an integer from 0 to 12;

j''' is 0, 1, or 2, wherein j''' is 0 when z'' is 0;

$X^{o'}$ and $X^{o'}$ each, independently, comprise a hydrophobic component;

b' is an integer from 0 to 12; and

10 b'' is an integer from 0 to 12.

287. The EEV of claim 286, wherein x' is 2, z' is 12, z'' is 2, and $X^{o'}$ and $X^{o'}$ are each, independently Nal.

15 288. The EEV of claim 287, comprising:

Ac-PKKKRKV-PEG₂-K(cyclo[FGFGRGRQ])-PEG₁₂-Nal-PEG₂-Nal-OH.

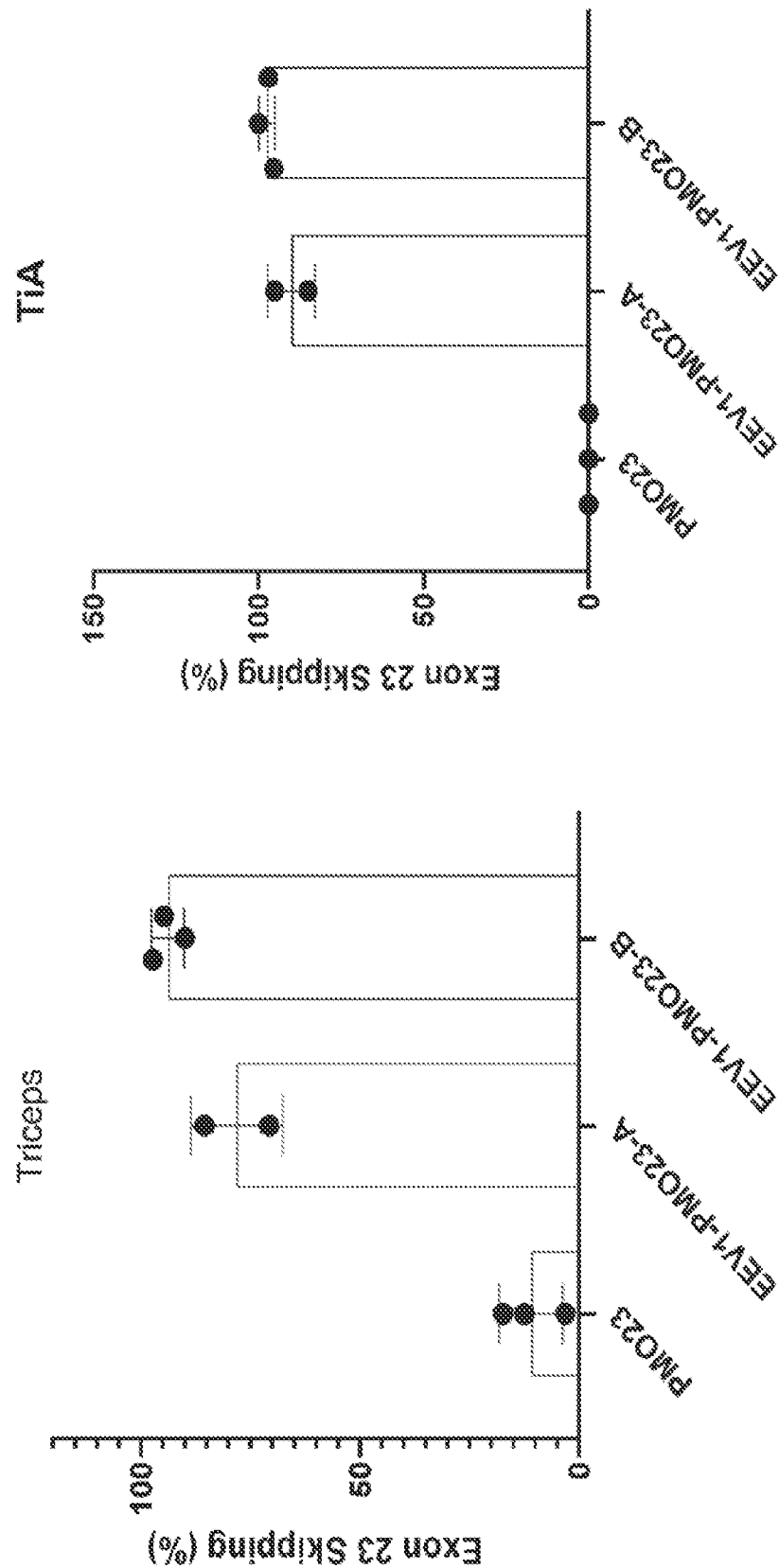


FIG. 1A

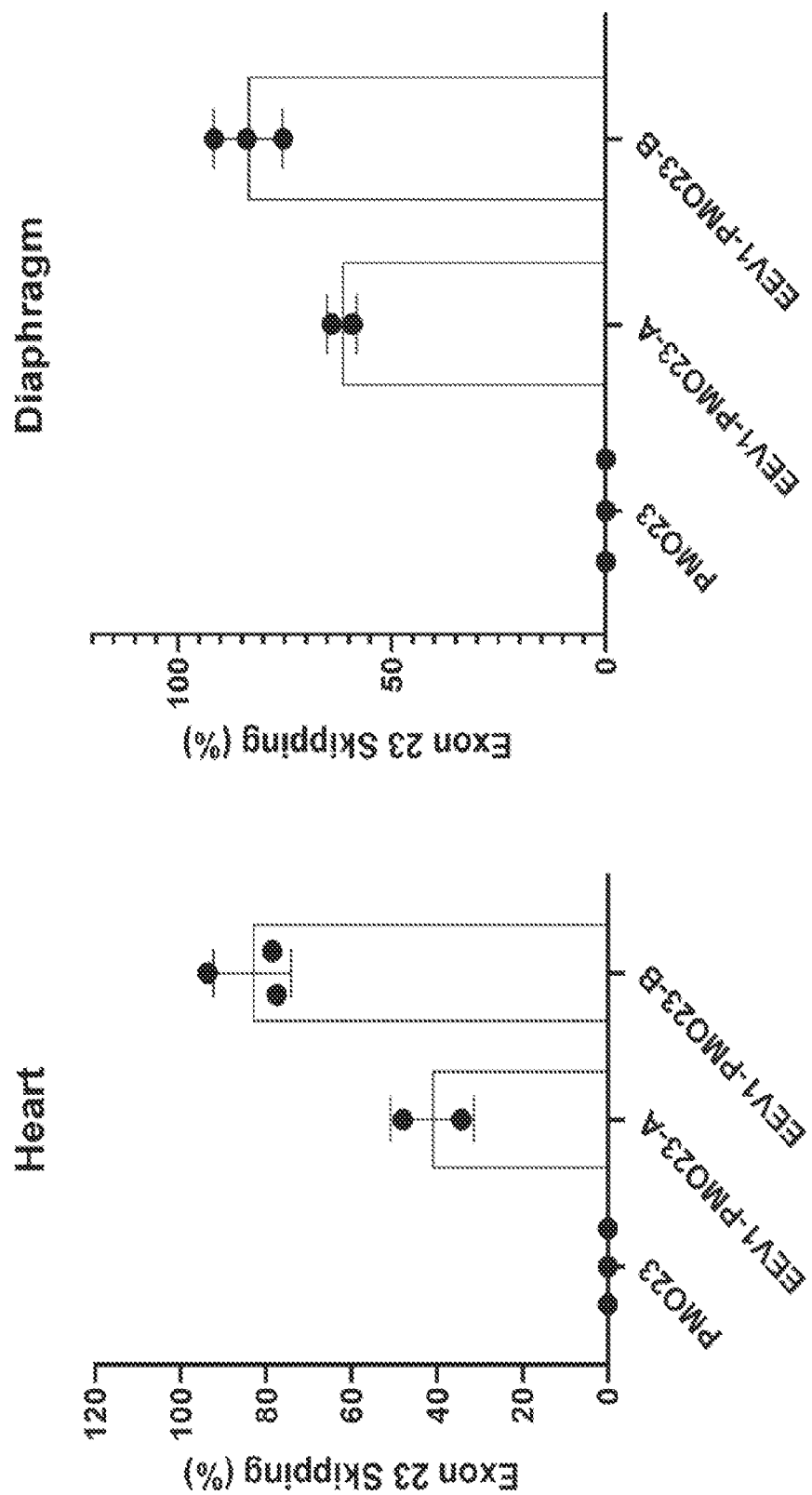
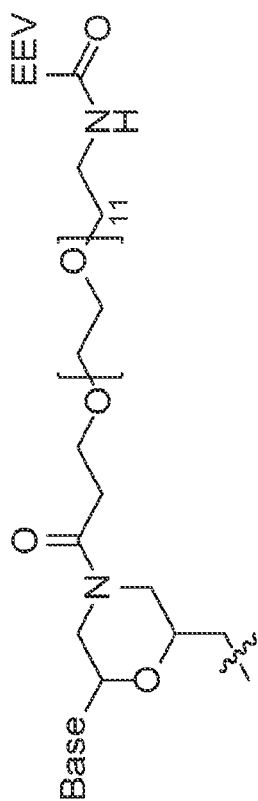


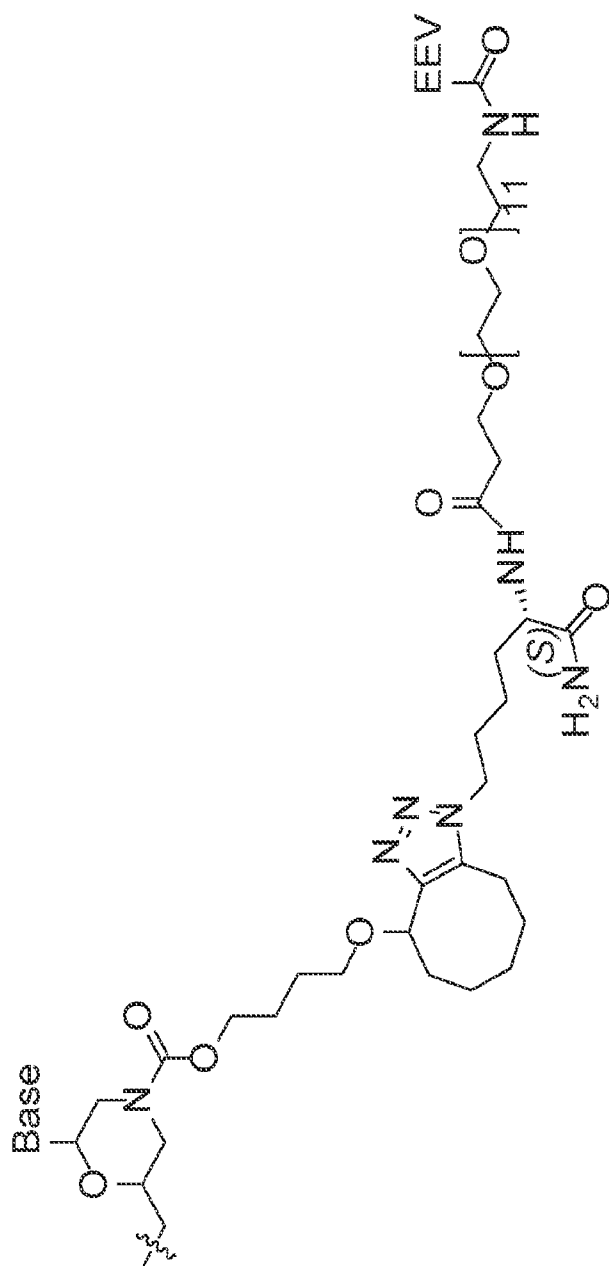
FIG. 1B



Linker for EEV1-PMO23-A
Installed using amide conjugation

PMO-23: GGCCAAACTCGGCTTACCTGAAAT

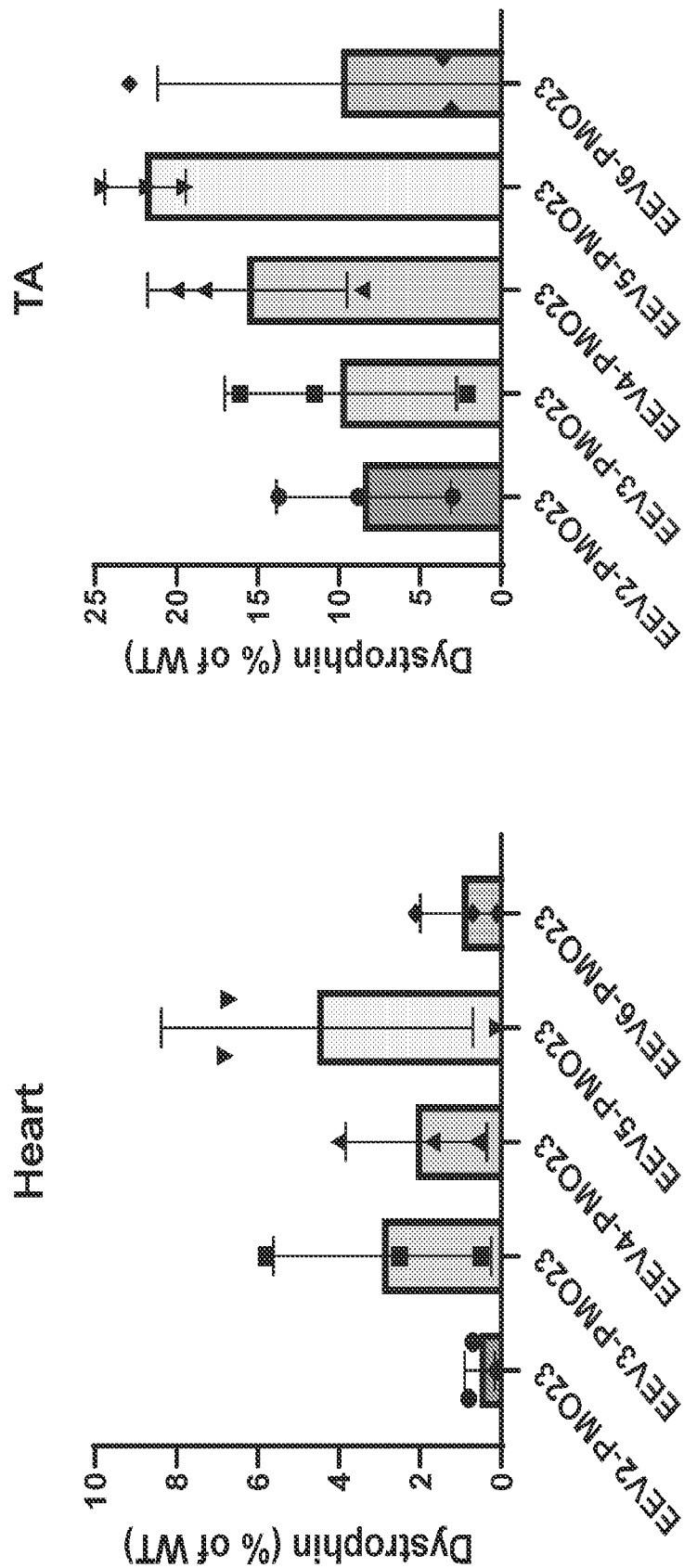
FIG. 1C



**Linker for EEV1-PMO23-B
Installed using strain-promoted
alkyne azide cycloaddition (SPAAC)**

EEV1: Ac-PPKKRKV-PEG₂-K(cyclo[F-Nal-Cit-r-Q])-Linker-PMO



2022 MDX-003 (AG): 7 d PI, 30 mpk (~40 mpk whole drug)

FIG. 2A

Peptide ID	Sequence
EEV2	Ac-PKKKRKV-PEG ₂ -K-(cyclo[(GfGGrQ)]-PEG ₁₂ -OH
EEV3	Ac-PKKKRKV-PEG ₂ -tetrazine-(cyclo[FGFGRGRQ])-PEG ₁₂ -OH
EEV4	Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(2-Nal)-OH
EEV5	Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -(2-Nal)-OH
EEV6	Ac-PKKKRKV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -C6-OH

FIG. 2B

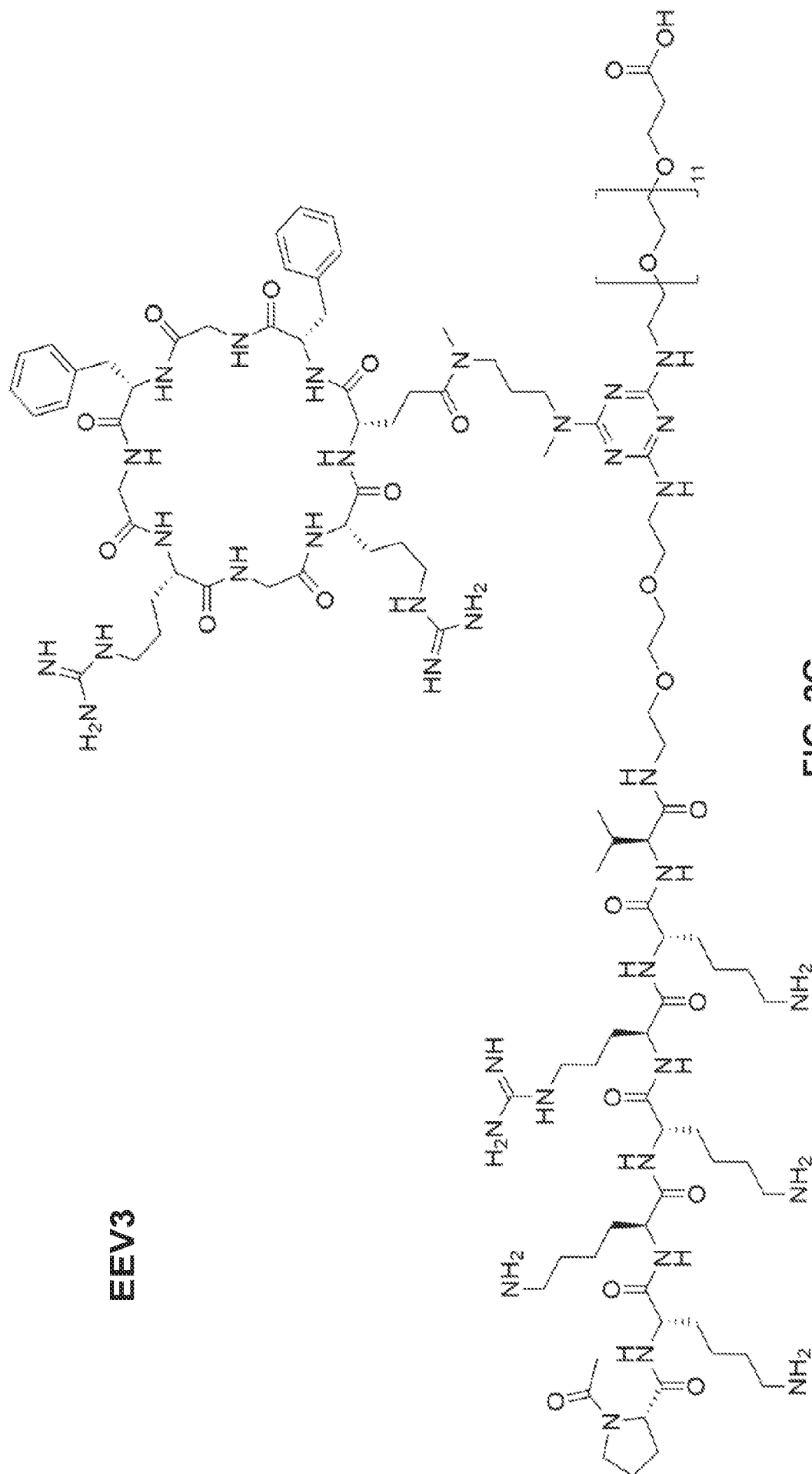


FIG. 2C

EEV3

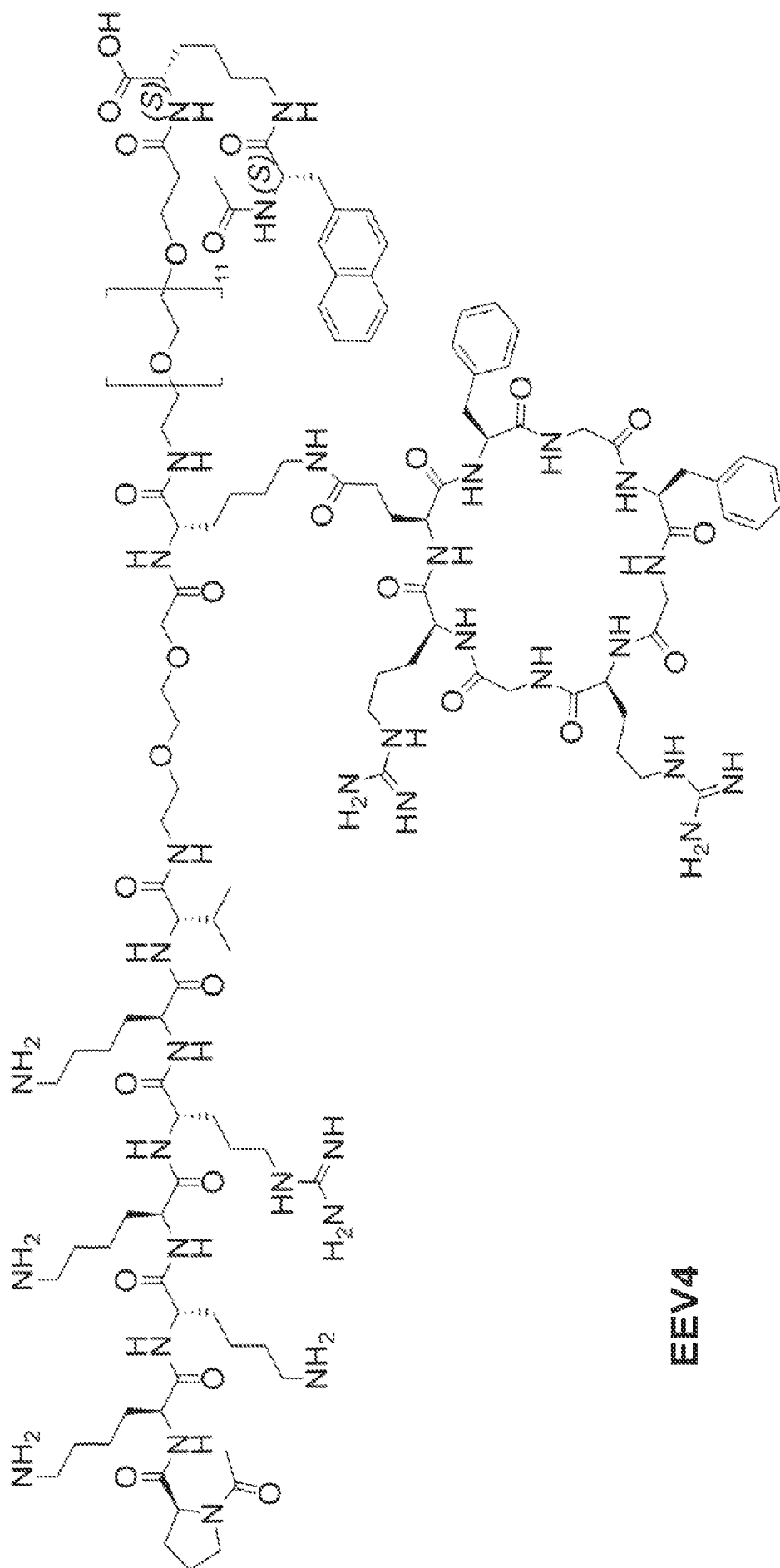
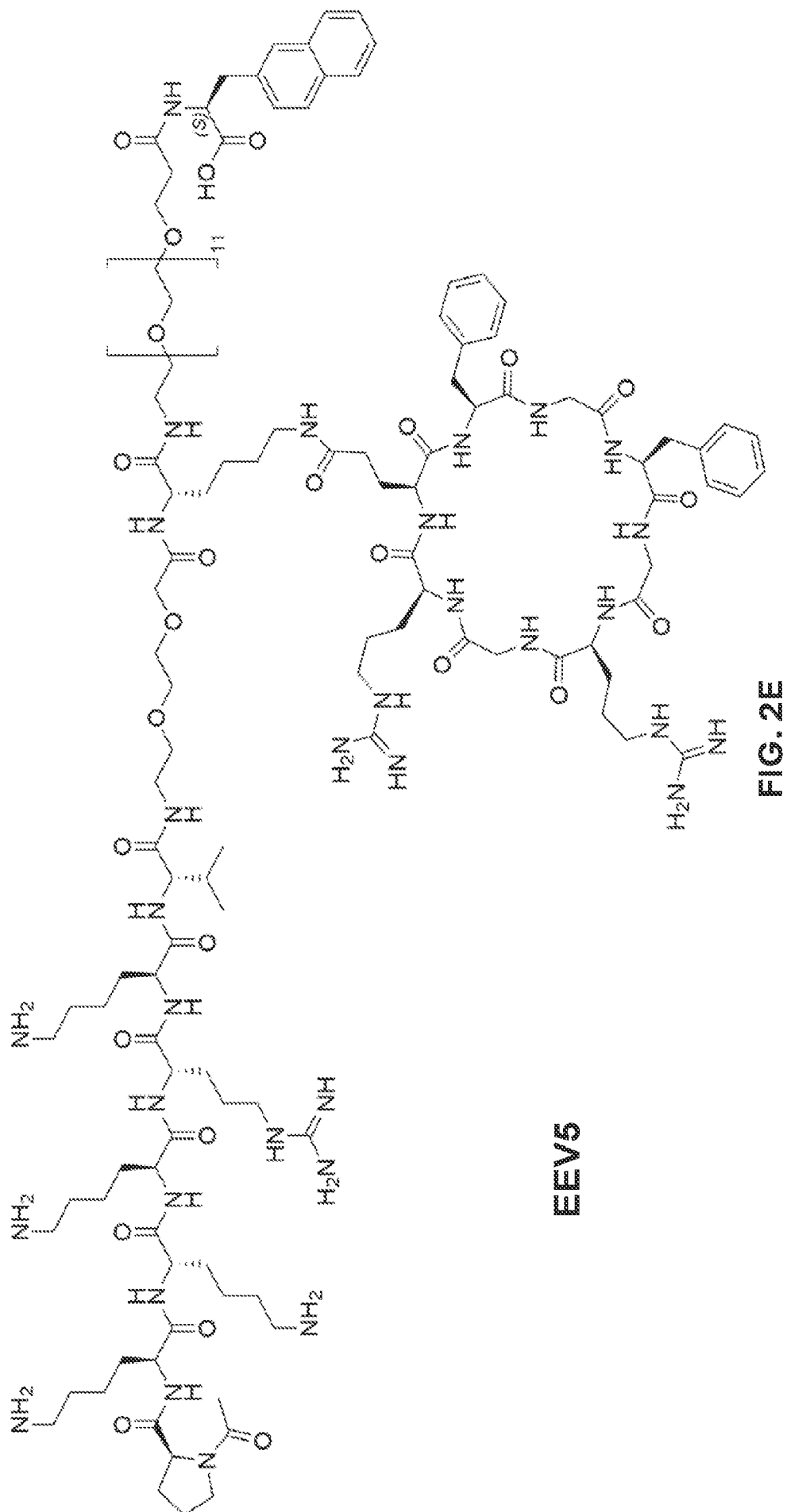


FIG. 2D

EEV4



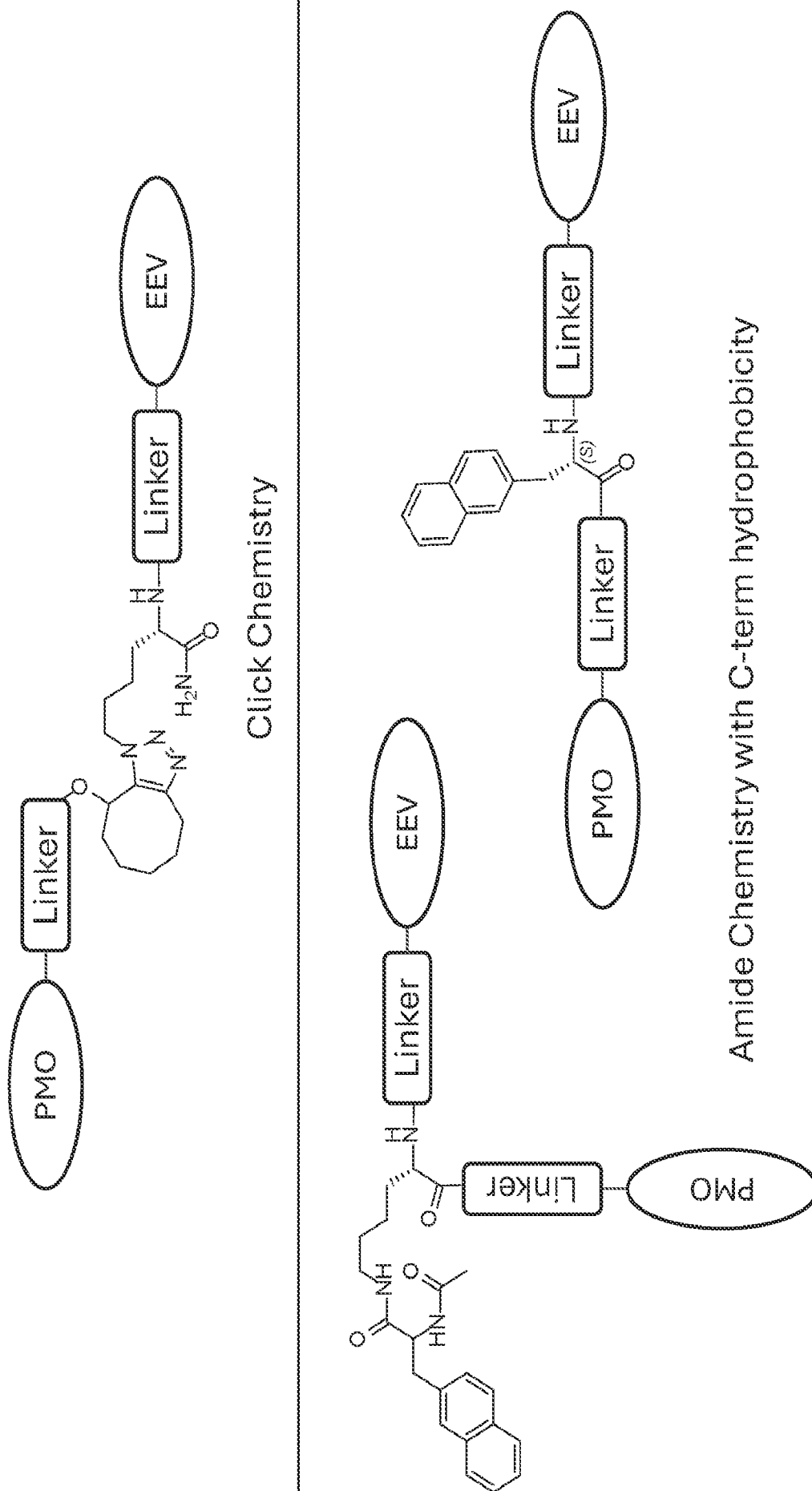
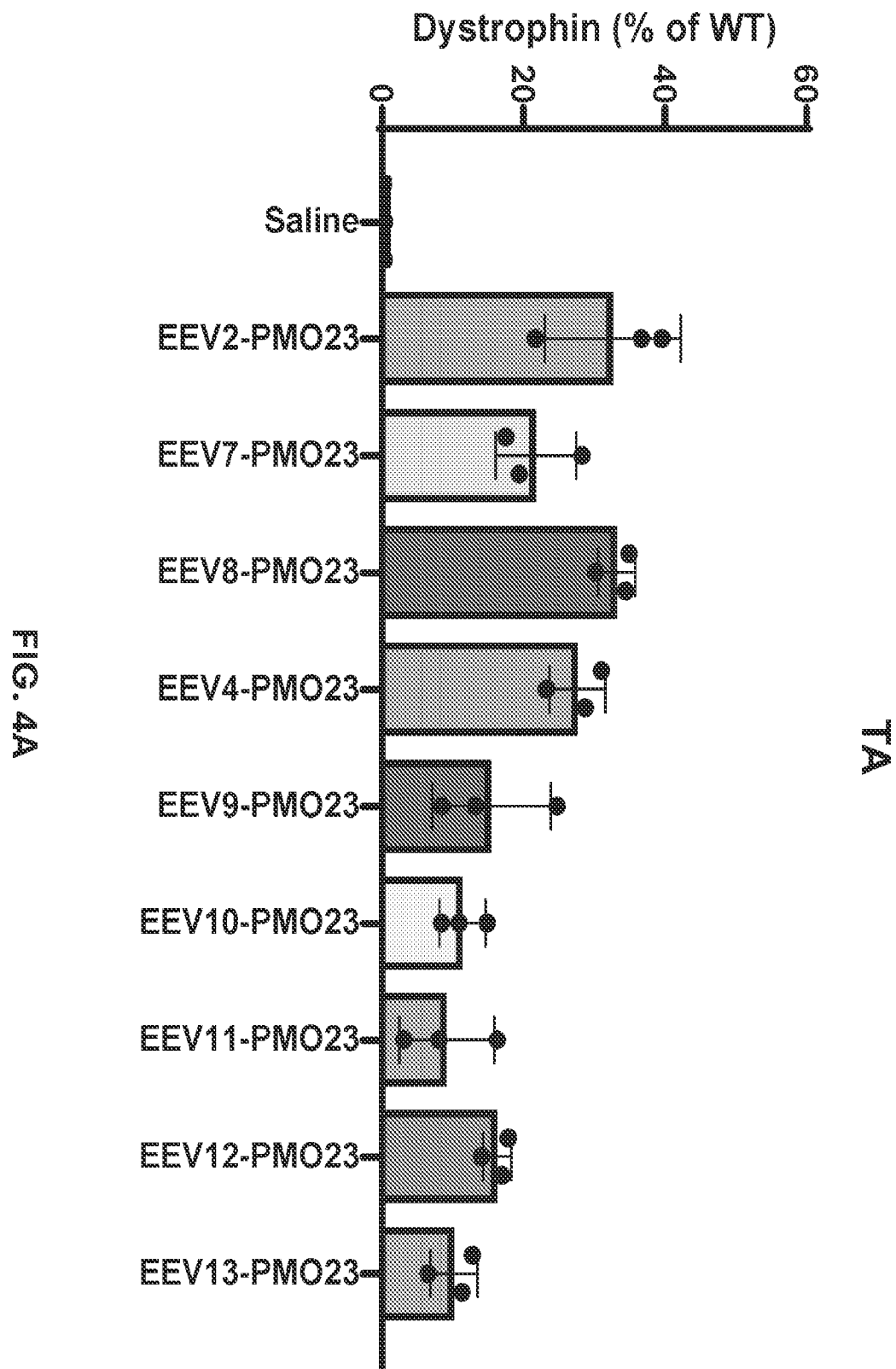
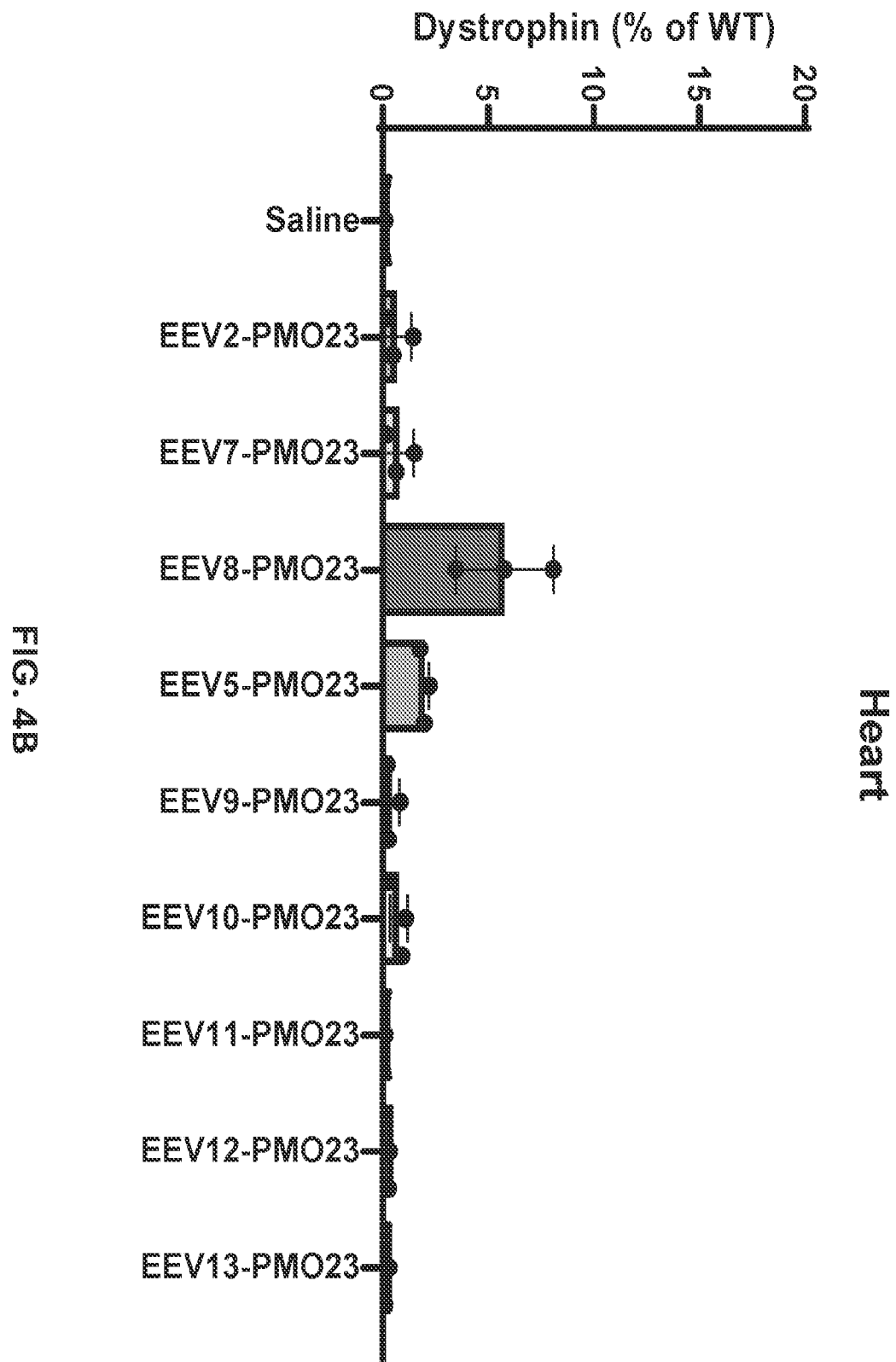


FIG. 3

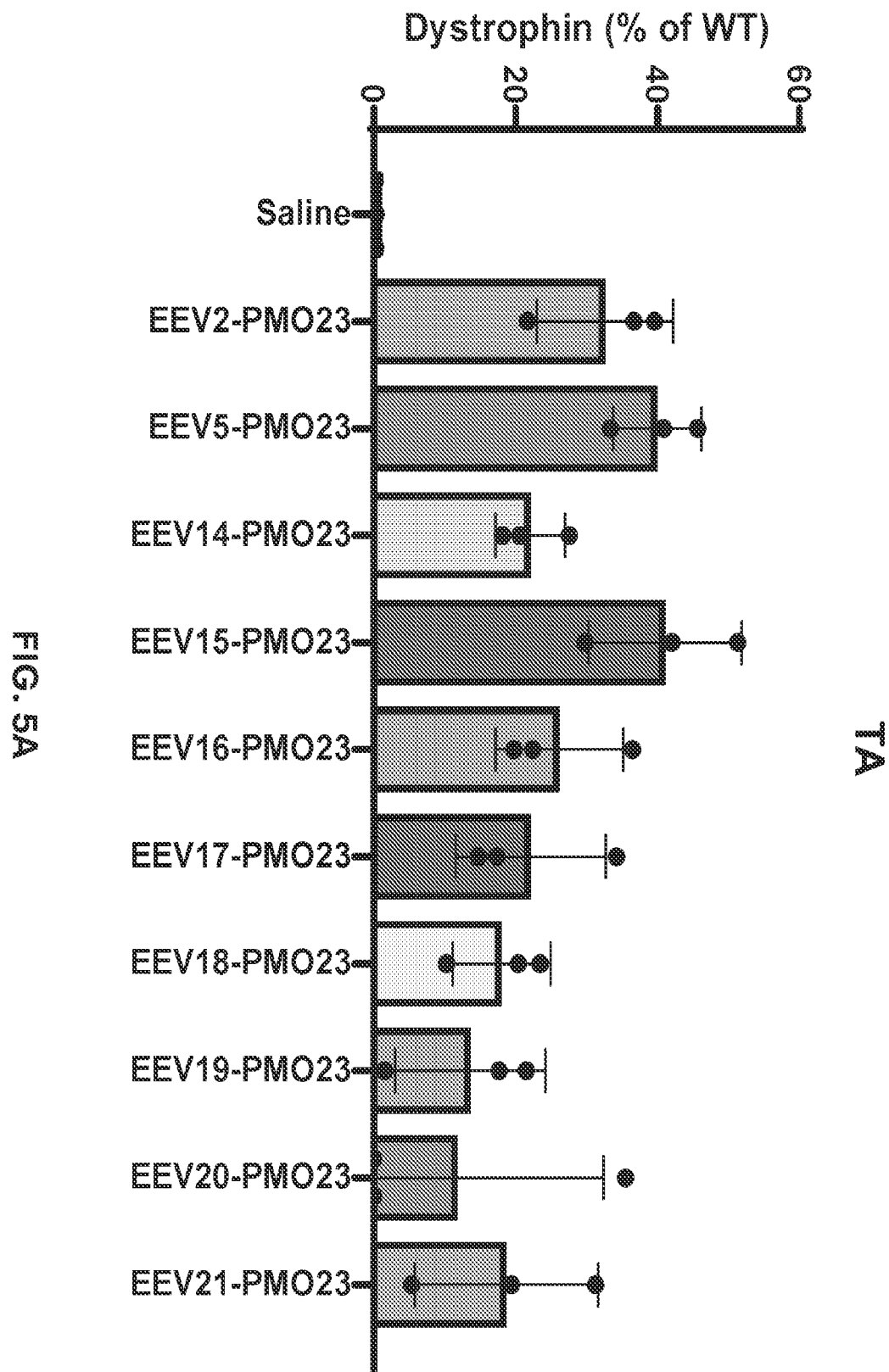


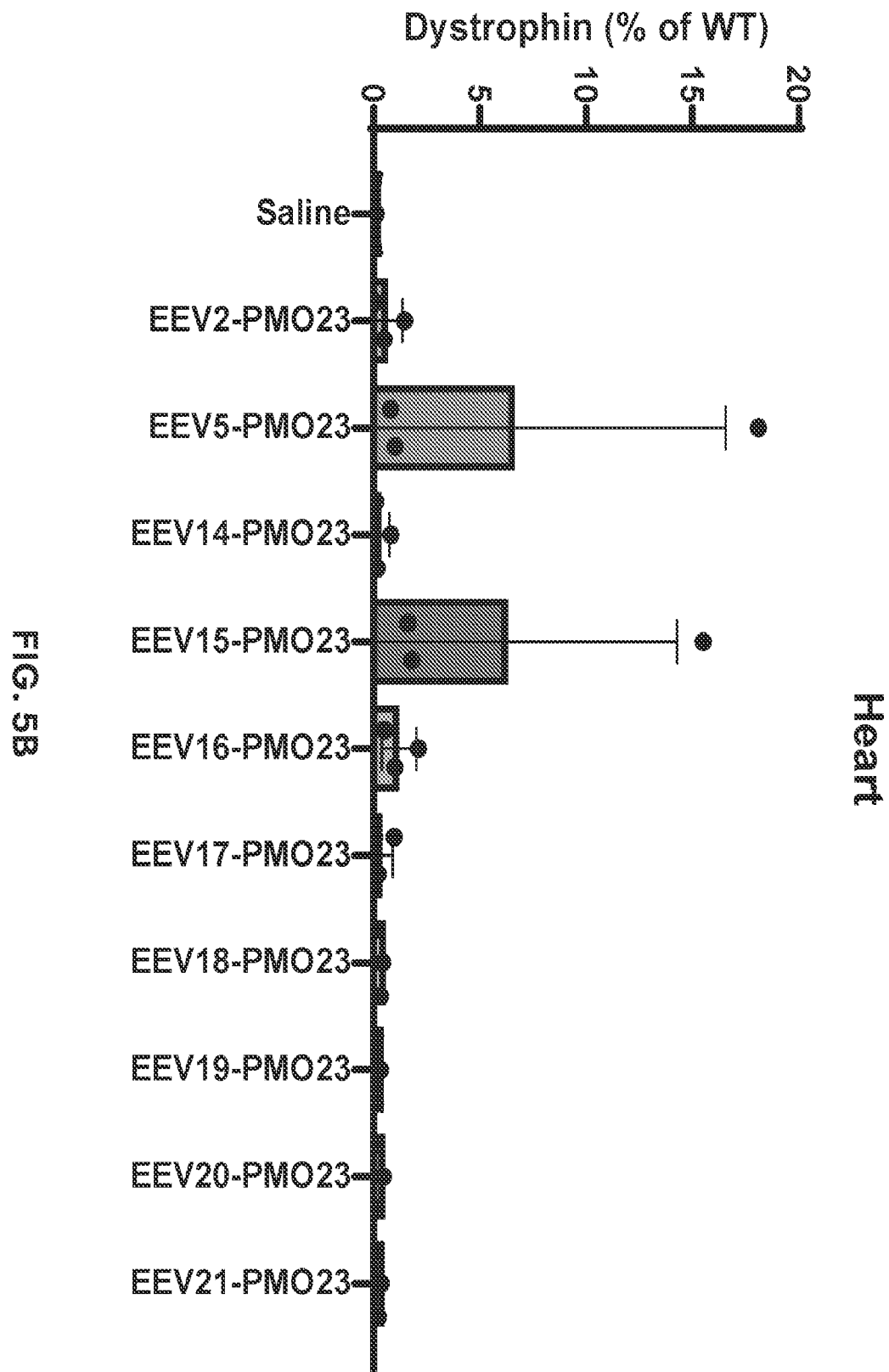


Name	Sequence
EEV2	Ac-PKKKRV-PEG ₂ -K-(cyclo[GfGfGrGrQ])-PEG ₁₂ -OH
EEV7	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(F)-OH
EEV8	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Bip)-OH
EEV4	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Nal)-OH
EEV9	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(W)-OH
EEV10	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(Y)-OH
EEV11	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(I)-OH
EEV12	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(L)-OH
EEV13	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRGRQ])-PEG ₁₂ -K(H)-OH

Bip = 4-biphenyl-L-alanine
All ε-functionalized Lys residues (e.g. N^ε-(N^ω-acetyl-L-leucyl)-L-lysine); see structure **EEV4** in FIG. 2D.

FIG. 4C





Name	Sequence
EEV2	Ac-PKKKRV-PEG ₂ -K-(cyclo[(GfGGrQ)]-PEG12-OH
EEV5	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-Nal-OH
EEV14	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-F-OH
EEV15	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-Bip-OH
EEV16	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-W-OH
EEV17	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-Y-OH
EEV18	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-V-OH
EEV19	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-I-OH
EEV20	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-L-OH
EEV21	Ac-PKKKRV-PEG ₂ -K(cyclo[FGFGRQ])-PEG12-H-OH

Bip = 4-biphenyl-L-alanine
See structure **EEV5** on FIG. 2E as an example

FIG. 5C

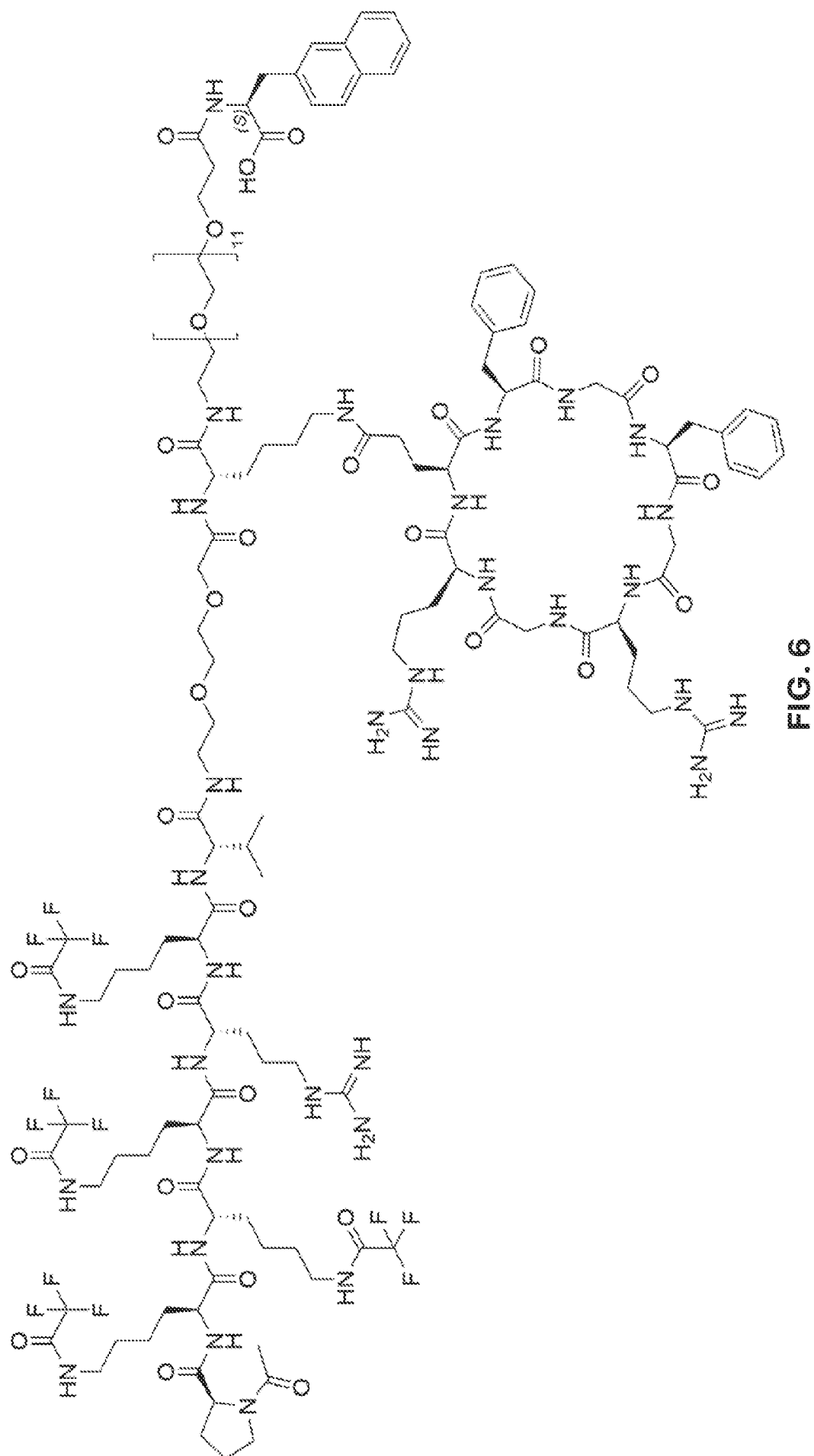
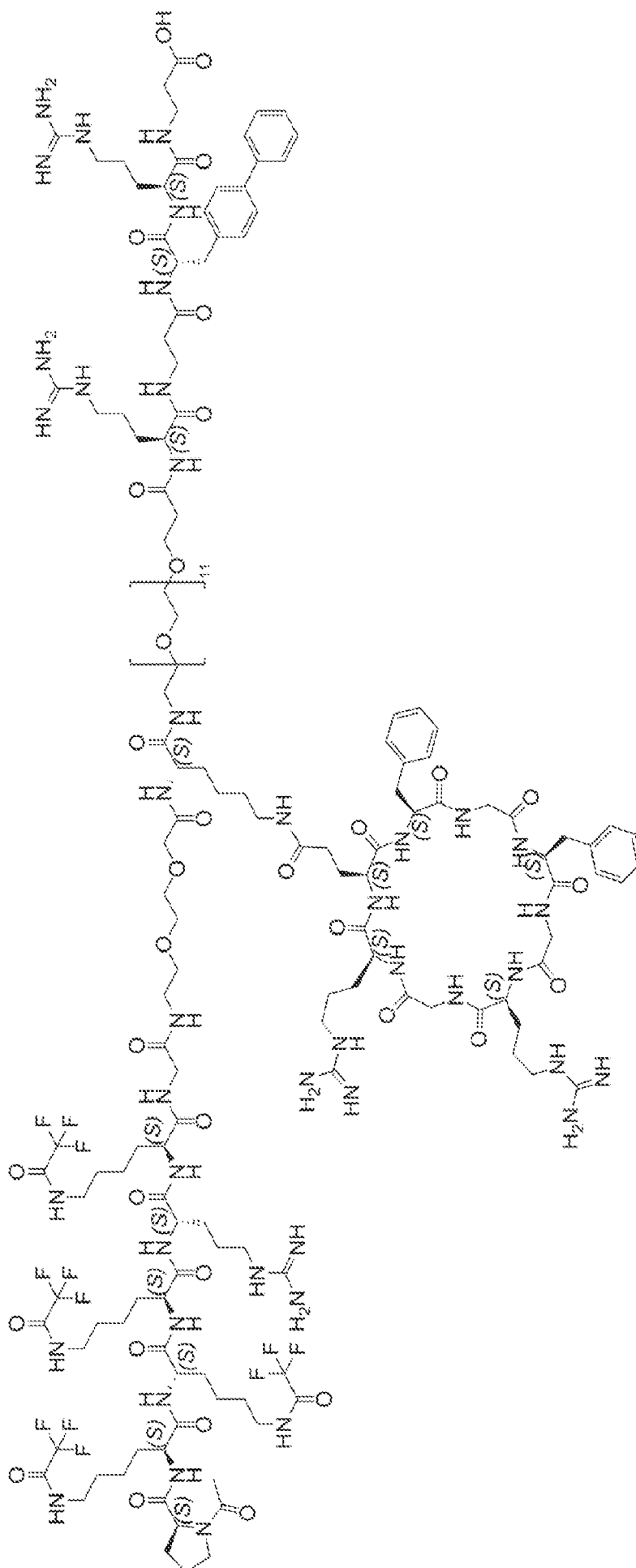


FIG. 6



LE

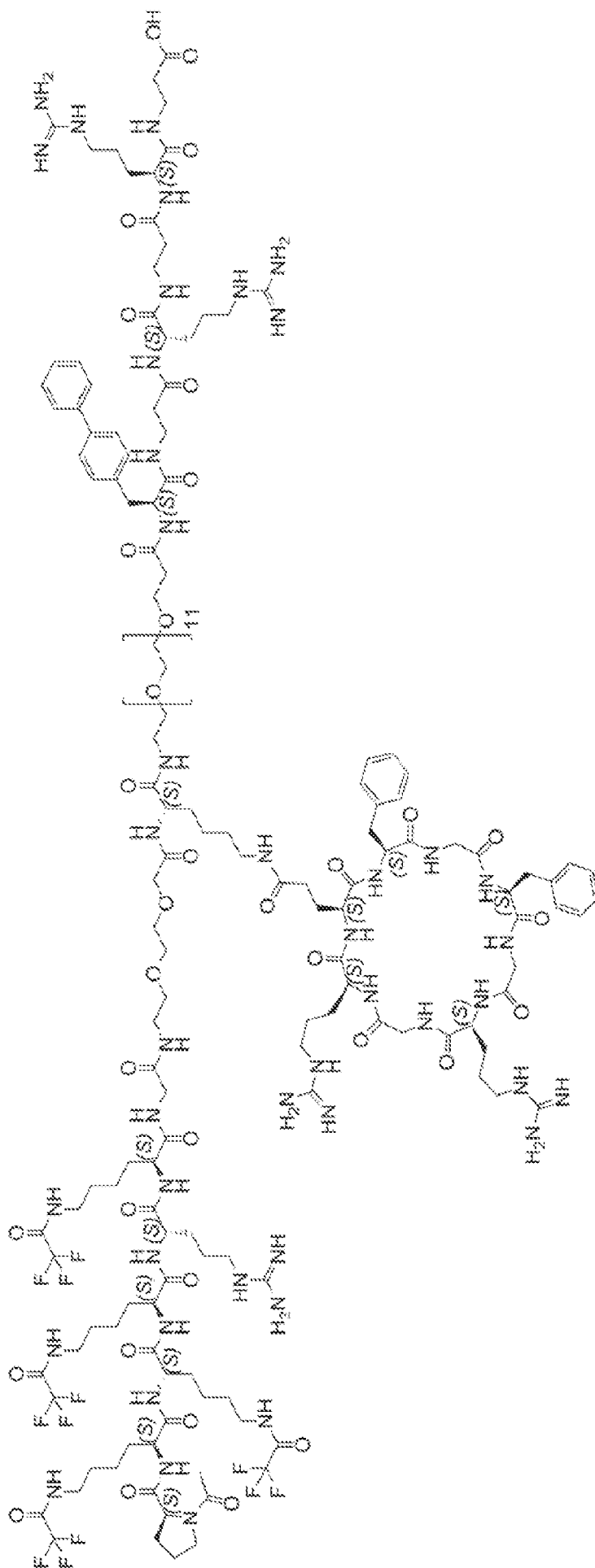


FIG. 8

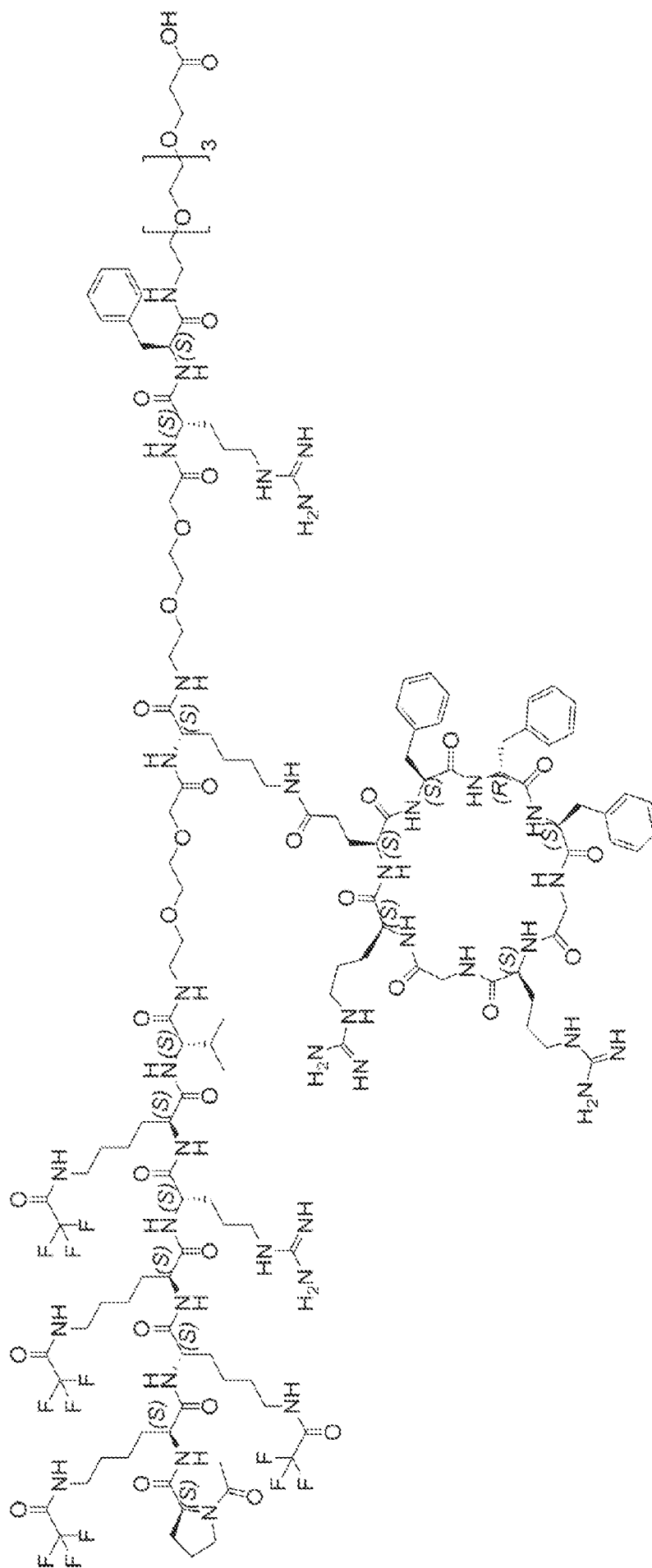


FIG. 9

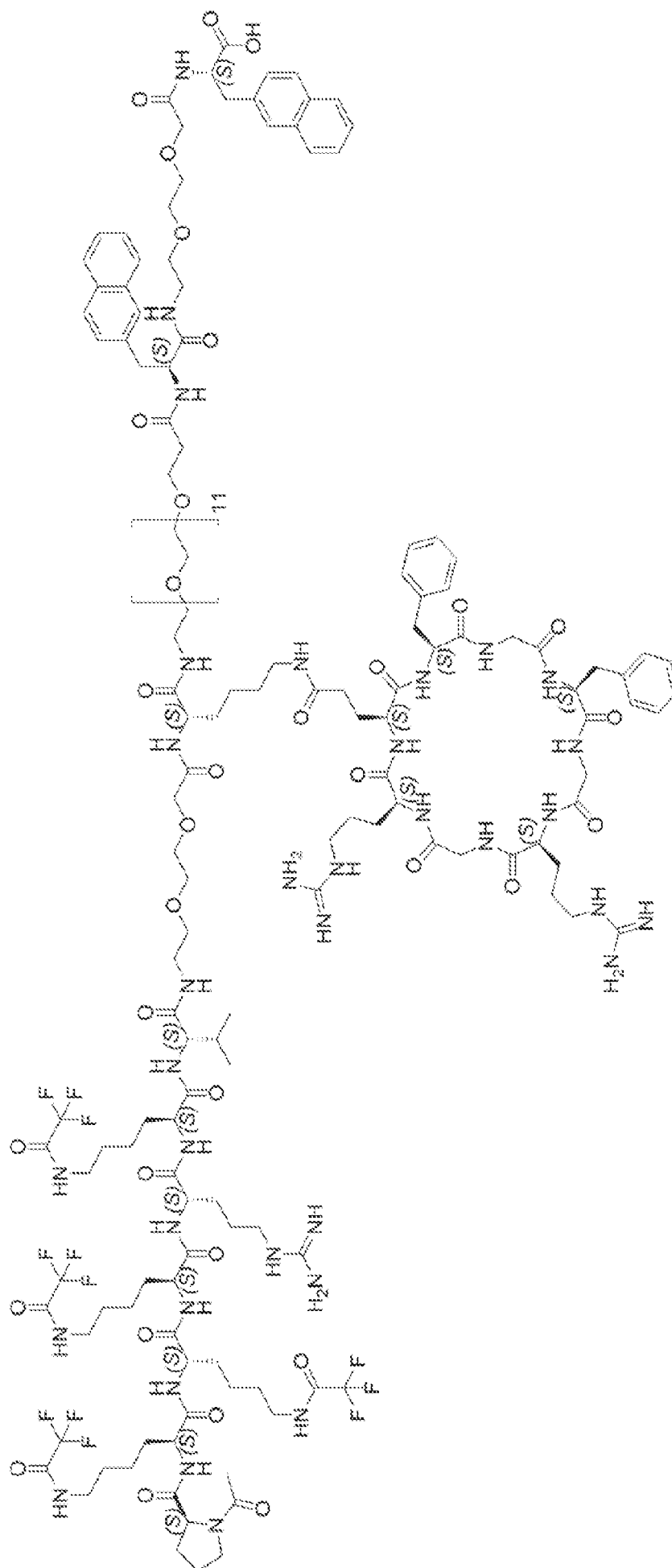


FIG. 10

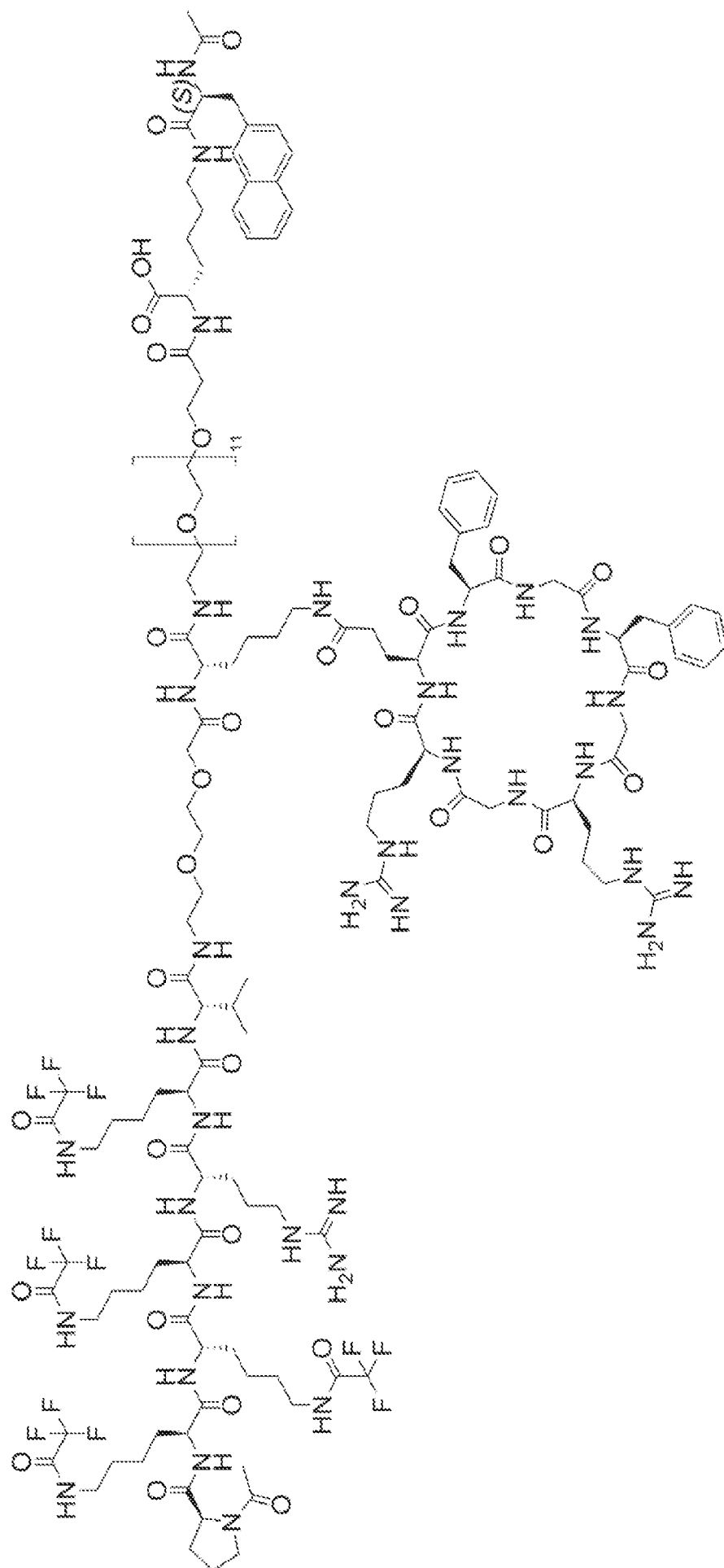


FIG. 11

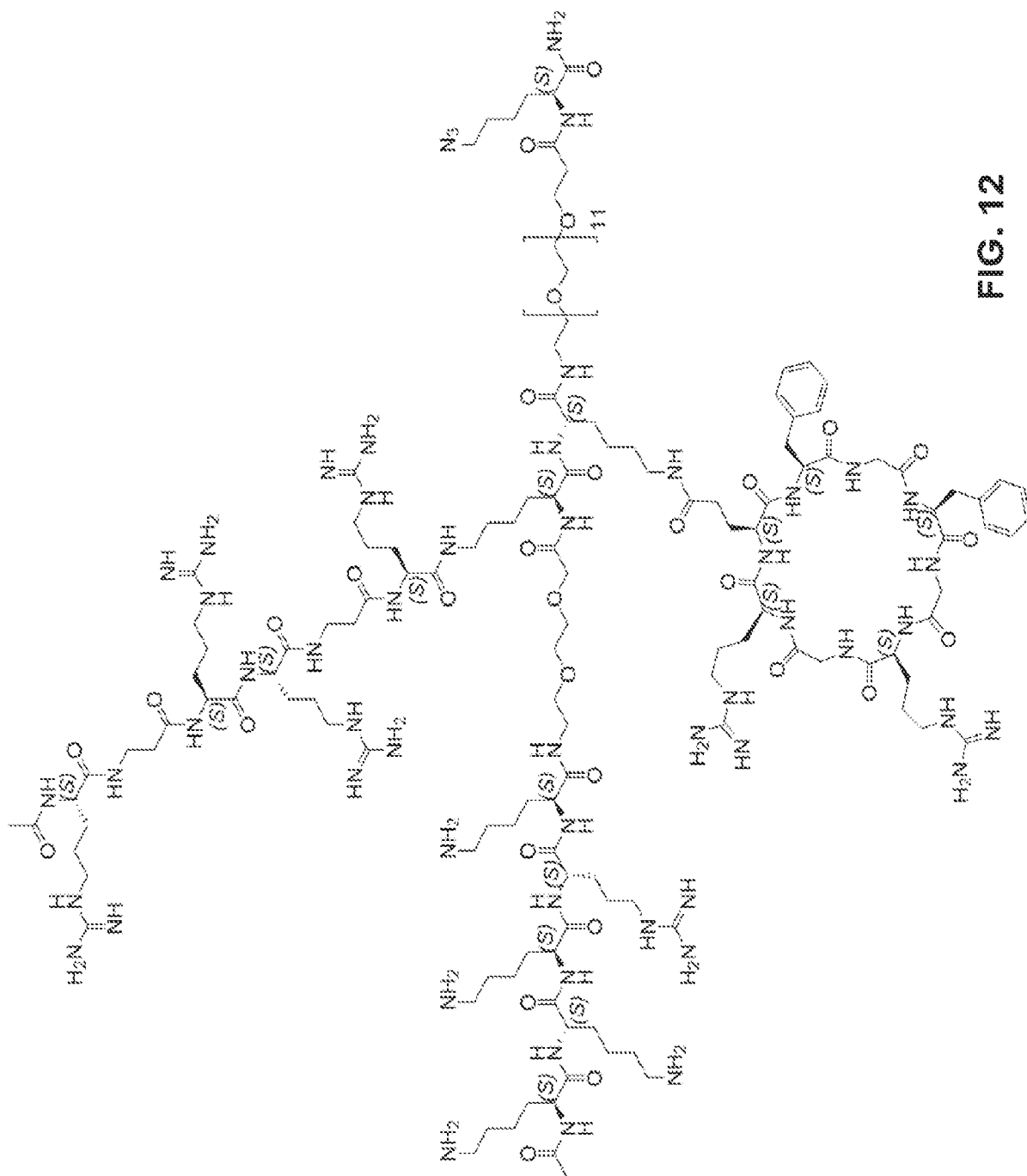


FIG. 12