# DE NOVO DESIGN OF POTENT AND SELECTIVE INTERLEUKIN MIMETICS

## Cross Reference

This application claims priority to U.S. Provisional Patent Application Serial Nos. 62/689,769 filed June 25, 2018 and 62/768,733 filed November 16, 2018, each incorporated by reference herein in its entirety.

## Background

The considerable potential of central immune cytokine interleukins such as IL-2 and IL-4 for cancer treatment has sparked numerous efforts to improve their therapeutic properties by mutation and/or chemical modification. However, because these approaches are closely tied to native IL-2 or IL-4, they cannot eliminate undesirable properties such as low stability and binding to the IL-2 receptor α subunit (IL-2Rα), to IL-4 receptor $\alpha\gamma_c$heterodimer (IL-4R$\alpha\gamma_c$), or to IL-13 receptor α subunit (IL-13Rα).

## Summary

In one example, a method is provided. A computing device determines a structure for a plurality of residues of a protein where the structure of the plurality of residues provides a particular receptor binding interface. The computing device determines a plurality of designed residues using a mimetic design protocol provided by the computing device, wherein the plurality of designed residues provide the particular receptor binding interface, and wherein the plurality of designed residues differ from the plurality of residues.

The computing device determines one or more connecting helix structures that connect the plurality of designed residues. The computing device determines a first protein backbone for the protein by assembling the one or more connecting helix structures and the plurality of designed residues over a plurality of combinations. The computing device designs a second protein backbone for the protein for flexibility and low energy structures based on the first protein backbone. The computing device generates an output related to at least the second protein backbone.

Also included are non-naturally occurring proteins prepared by the methods described herein. The non-naturally occurring proteins can be cytokines, for example, non-naturally occurring IL-2 or IL-4 (also referred to herein as IL-2, IL-2/15 mimetics or IL-4 mimetics).

In another example, a computing device is provided. The computing device includes one or more processors; and data storage that is configured to store at least computer-readable instructions that, when executed by the one or more processors, cause the computing device to perform functions. The functions include: determining a structure for a plurality of residues of a protein that provides a particular receptor binding interface; determining a plurality of designed residues using a mimetic design protocol, wherein the plurality of designed residues provide the particular receptor binding interface, and wherein the plurality of designed residues differ from the plurality of residues; determining one or more connecting helix structures that connect the plurality of designed residues; determining a first protein backbone for the protein by assembling the one or more connecting helix structures and the plurality of designed residues over a plurality of combinations; designing a second protein backbone for the protein for flexibility and low energy structures based on the first protein backbone; and generating an output related to at least the second protein backbone for the protein.

In another example, a non-transitory computer-readable medium is provided. The non-transitory computer-readable medium is configured to store at least computer-readable instructions that, when executed by one or more processors of a computing device, cause the computing device to perform functions. The functions include: determining a structure for a plurality of residues of a protein that provides a particular receptor binding interface; determining a plurality of designed residues using a mimetic design protocol, wherein the plurality of designed residues provide the particular receptor binding interface, and wherein the plurality of designed residues differ from the plurality of residues; determining one or more connecting helix structures that connect the plurality of designed residues; determining a first protein backbone for the protein by assembling the one or more connecting helix structures and the plurality of designed residues over a plurality of combinations; designing a second protein backbone for the protein for flexibility and low energy structures based on the first protein backbone; and generating an output related to at least the second protein backbone for the protein.

In another example, a device is provided. The device includes: means for determining a structure for a plurality of residues of a protein that provides a particular receptor binding interface; means for determining a plurality of designed residues using a mimetic design protocol, wherein the plurality of designed residues provide the particular receptor binding

interface, and wherein the plurality of designed residues differ from the plurality of residues; means for determining one or more connecting helix structures that connect the plurality of designed residues; means for determining a first protein backbone for the protein by assembling the one or more connecting helix structures and the plurality of designed residues over a plurality of combinations; means for designing a second protein backbone for the protein for flexibility and low energy structures based on the first protein backbone; and means for generating an output related to at least the second protein backbone for the protein.

In another example, non-naturally occurring polypeptides are provided comprising domains X1, X2, X3, and X4, wherein:

(a) X1 is a peptide comprising the amino acid sequence at least 25%, 27%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98%, or 100% identical to **E**HA**LYD**A**L** (SEQ ID NO:1);

(b) X2 is a helical-peptide of at least 8 amino acids in length;

(c) X3 is a peptide comprising the amino acid sequence at least 25%%, 27%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98%, or 100% identical to **Y**AF**N**FE**LI** (SEQ ID NO:2);

(d) X4 is a peptide comprising the amino acid sequence at least 25%, 27%, 30%, 35%, 40%, 45%, 50%, 55%, 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%, 98%, or 100% identical to **I**TIL**Q**S**W**IF (SEQ ID NO:3);

wherein X1, X2, X3, and X4 may be in any order in the polypeptide;

wherein amino acid linkers may be present between any of the domains; and

wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), IL-4 receptor $\alpha\gamma_c$ heterodimer (IL-4R$\alpha\gamma_c$), or IL-13 receptor α subunit (IL-13Rα).

In one aspect, the present disclosure refers to a non-naturally occurring polypeptide comprising domains X1, X2, X3, and X4, wherein:

X1 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide PKKKIQLHAEHALYDALMILNI (SEQ ID NO: 4);

X2 is a helical-peptide of at least 8 amino acids in length;

X3 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide LEDYAFNFELILEEIARLFESG (SEQ ID NO:5); and

X4 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide EDEQEEMANAIITILQSWIFS (SEQ ID NO:6),

wherein X1, X2, X3, and X4 may be in any order in the polypeptide;

wherein amino acid linkers may be present between any of the domains; and

wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$) or IL-4 receptor $\alpha\gamma_c$heterodimer (IL-4R$\alpha\gamma_c$).

In one aspect, the present disclosure refers to a non-naturally occurring polypeptide comprising domains X1, X2, X3, and X4, wherein:

(a) X1 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to EHALYDAL (SEQ ID NO:1);

(b) X2 is a helical-peptide of at least 8 amino acids in length;

(c) X3 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to YAFNFELI (SEQ ID NO:2);

(d) X4 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to ITILQSWIF (SEQ ID NO:3);

wherein X1, X2, X3, and X4 may be in any order in the polypeptide;

wherein amino acid linkers may be present between any of the domains; and

wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

In one aspect, the present disclosure refers to a non-naturally occurring polypeptide comprising an amino acid sequence at least 90%, at least 95%, at least 98% or 100% identical to the amino acid sequence set forth in SEQ ID NO: 90 wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

In one aspect, the present disclosure refers to a non-naturally occurring polypeptide comprising an amino acid sequence at least 90%, at least 95%, at least 98% or 100% identical to the amino acid sequence set forth in SEQ ID NO: 181 wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

In one aspect, the present disclosure refers to a recombinant nucleic acid encoding any one of the polypeptides disclosed herein.

In one aspect, the present disclosure refers to a recombinant host cell comprising the nucleic acid disclosed herein.

In one aspect, the present disclosure refers to a pharmaceutical composition comprising any one of the polypeptides disclosed herein that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$) or a recombinant nucleic acid disclosed herein that encodes a polypeptide that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), and a pharmaceutically acceptable carrier.

In one aspect, the present disclosure refers to use of any one of the polypeptides disclosed herein that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), a recombinant nucleic acid disclosed herein that encodes a polypeptide that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), or the pharmaceutical composition disclosed herein in the manufacture of a medicament for treating cancer that expresses IL-2 receptor $\beta\gamma$c heterodimer (IL-2R$\beta\gamma$c) in a subject having said cancer.

In other examples are provided pharmaceutical compositions comprising one or more polypeptide disclosed herein and a pharmaceutically acceptable carrier, recombinant nucleic acids encoding a polypeptide disclosed herein, expression vectors comprising the recombinant nucleic acids disclosed herein, and recombinant host cells comprising one or more expression vector disclosed herein. In a further example, methods for treating cancer are provided, comprising administering to a subject having cancer one or more polypeptide, recombinant nucleic acid, expression vector comprising the recombinant nucleic acid, and/or recombinant host cells disclosed herein or a pharmaceutical composition thereof in an amount effective to treat the tumor.

**We claim**

1. A non-naturally occurring polypeptide comprising domains X1, X2, X3, and X4, wherein:

X1 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide PKKKIQLHAEHALYDALMILNI (SEQ ID NO: 4);

X2 is a helical-peptide of at least 8 amino acids in length;

X3 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide LEDYAFNFELILEEIARLFESG (SEQ ID NO:5); and

X4 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide EDEQEEMANAIITILQSWIFS (SEQ ID NO:6),

wherein X1, X2, X3, and X4 may be in any order in the polypeptide;

wherein amino acid linkers may be present between any of the domains; and

wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$) or IL-4 receptor $\alpha\gamma_c$heterodimer (IL-4R$\alpha\gamma_c$).

2. The polypeptide of claim 1, provided that:

(a) X1 comprises an amino acid sequence at least 25%, at least 27%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to EHALYDAL (SEQ ID NO:1);

(b) X3 comprises an amino acid sequence at least 25%, at least 27%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to YAFNFELI (SEQ ID NO:2); and

(c) X4 comprises an amino acid sequence at least 25%, at least 27%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to ITILQSWIF (SEQ ID NO:3).

3. A non-naturally occurring polypeptide comprising domains X1, X2, X3, and X4, wherein:

(a) X1 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to EHALYDAL (SEQ ID NO:1);

(b) X2 is a helical-peptide of at least 8 amino acids in length;

(c) X3 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to YAFNFELI (SEQ ID NO:2);

(d) X4 is a peptide comprising an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to ITILQSWIF (SEQ ID NO:3);

wherein X1, X2, X3, and X4 may be in any order in the polypeptide;

wherein amino acid linkers may be present between any of the domains; and

wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

4. The polypeptide of claim 2 or 3, wherein:

(i) X1 includes both of the following: H at residue 2 and Y at residue 5 relative to SEQ ID NO:1; and/or

(ii) X3 includes all 5 of the following: Y at residue 1, F at residue 3, N at residue 4, L at residue 7, and I at residue 8 relative to SEQ ID NO:2; wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

5. The polypeptide of any one of claims 1-3 wherein,

X1 comprises the amino acid sequence EHALYDAL (SEQ ID NO:1);

X3 is comprises the amino acid sequence YAFNFELI (SEQ ID NO:2); and

X4 comprises the amino acid sequence ITILQSWIF (SEQ ID NO:3); wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

6. The polypeptide of any one of claims 1 - 5, wherein:

X1 comprises an amino acid sequence at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide PKKKIQLHAEHALYDALMILNI (SEQ ID NO: 4);

X3 comprises an amino acid sequence at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide LEDYAFNFELILEEIARLFESG (SEQ

ID NO:5); and

X4 comprises an amino acid at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide EDEQEEMANAIITILQSWIFS (SEQ ID NO:6); wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

7. The polypeptide of any one of claims 1-2 or 6, wherein:

(i) X1 includes all 5 of the following: L at residue 7, H at residue 8, H at residue 11, Y at residue 14; M at residue 18 relative to SEQ ID NO:4; and/or

(ii) X3 includes all 8 of the following: D at residue 3, Y at residue 4, F at residue 6, N at residue 7, L at residue 10, I at residue 11, E at residue 13, or E at residue 14 relative to SEQ ID NO:5; wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

8. The polypeptide of any one of claims 1 or 2, wherein:

X1 comprises an amino acid sequence at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide PKKKIQIMAEEALKDALSILNI (SEQ ID NO: 8);

X3 comprises an amino acid sequence at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide LERFAKRFERNLWGIARLFESG (SEQ ID NO: 9); and

X4 comprises an amino acid sequence at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide EDEQEEMANAIITILQSWFFS (SEQ ID NO: 10).

wherein

(i) X1 includes I at residue 7, T or M at residue 8, E at residue 11, K at residue 14 and S at residue 18 relative to SEQ ID NO:8; and

(ii) X3 includes R at residue 3, F at residue 4, K at residue 6, R at residue 7, R at residue 10, N at residue 11, W at residue 13, and G at residue 14 relative to SEQ ID NO:9, wherein the polypeptide binds to IL-4 receptor $\alpha\gamma_c$heterodimer (IL-4R$\alpha\gamma_c$).

9. The polypeptide of any one of claims 1-2 or 6-7, wherein amino acid substitutions relative to the reference peptide domains do not occur at positions 10, 13, 14, 15, and 17 relative to SEQ ID NO:4; positions 1, 4, 7, 10, 11, and 15 relative to SEQ ID NO:5; and

positions 12, 16, and 18 relative to SEQ ID NO:6.

10. The polypeptide of any one of claims 1-9, wherein amino acid substitutions relative to the reference peptide domains are conservative amino acid substitutions.

11. The polypeptide of any one of claims 1-10, wherein the domains are arranged N-terminal to C-terminal in an arrangement selected from X1-X3-X2-X4.

12. The polypeptide of any one of claims 1-11, wherein X2 is a peptide comprising an amino acid sequence at least 25%, at least 27%, at least 30%, at least 35%, at least 40%, at least 45%, at least 50%, at least 55%, at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to KDEAEKAKRMKEWMKRIKT (SEQ ID NO:7).

13. The polypeptide of claim 12, wherein X2 comprises an amino acid sequence at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to KDEAEKAKRMKEWMKRIKT (SEQ ID NO:7).

14. The polypeptide of claim 1 or 2, wherein

X1 comprises an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide PKKKIQLHAEHALYDALMILNI (SEQ ID NO: 4) wherein amino acid residues relative to SEQ ID NO:4 are selected from:

| | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|
| Position 01: | A | F | I | L | M | P | Q | R | S | W |
| Position 02: | A | D | E | G | V | K | | | | |
| Position 03: | D | E | F | W | K | | | | | |
| Position 04: | D | E | K | N | P | R | W | | | |
| Position 05: | D | E | H | I | K | L | M | S | | |
| Position 06: | A | D | E | G | L | P | S | W | Q | |
| Position 07: | D | E | L | Q | Y | I | | | | |
| Position 08: | A | F | H | W | Y | M | T | | | |
| Position 09: | C | F | P | A | | | | | | |
| Position 10: | C | D | E | F | K | P | | | | |
| Position 11: | D | F | H | E | | | | | | |

Position 12: A D E P S T V

Position 13: H I L M P R V W

Position 14: F R W Y K

Position 15: D E N Y

Position 16: A C L M S

Position 17: F I L M P R

Position 18: G M Q Y S

Position 19: I L M P Q V

Position 20: A K L M Q R S

Position 21: G K N P R S W

Position 22: D E I K M N W Y;

X3 comprises an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide LEDYAFNFELILEEIARLFESG (SEQ ID NO:5) wherein amino acid residues relative to SEQ ID NO:5 are selected from:

Position 01: A L

Position 02: D E G K M T

Position 03: D E N Y R

Position 04: C D G T Y F

Position 05: A F H S V W Y

Position 06: A F I M T V Y K

Position 07: D K N S T R

Position 08: A C G L M S V F

Position 09: C H K L R S T V E

Position 10: F I L M Y R

Position 11: I L N T Y

Position 12: F K L M S V

Position 13: A D F G I N P Q S T E W

Position 14: A E F G H S V

Position 15: C I L M V W

Position 16: A D G S T V

Position 17: H K L N R

Position 18: C D G I L Q R T W

Position 19: D F M N W

Position 20: A C E F G M S Y

Position 21: D E G H L M R S T V W

Position 22: A D G K N S Y;

wherein X4 comprises an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the peptide EDEQEEMANAIITILQSWIFS (SEQ ID NO:6) wherein amino acid residues relative to SEQ ID NO:6 are selected from:

Position 01: D E G K V

Position 02: D I M S

Position 03: E G H K

Position 04: E G I K Q R S

Position 05: A D E G H S V

Position 06: C D E G I M Q R T V

Position 07: C E L M P R T

Position 08: A F L M W

Position 09: A G L N Q R T

Position 10: A C D E F H I W

Position 11: I M N S V W

Position 12: I K L S V

Position 13: C L M R S T

Position 14: I L P T Y

Position 15: F G I L M N V

Position 16: H K Q R

Position 17: C F K S W Y

Position 18: K Q T W

Position 19: C G N I

Position 20: C F G L Y

Position 21: A F G H S Y; and

wherein X2 comprises an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to KDEAEKAKRMKEWMKRIKT (SEQ ID NO:7) wherein amino acid residues relative to SEQ ID NO:7 are selected from:

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Position 01: | A | H | L | M | R | S | V | K | | |
| Position 02: | A | D | E | Q | R | S | T | V | W | Y |
| Position 03: | C | E | G | K | L | N | Q | R | W | |
| Position 04: | A | F | G | N | S | T | V | Y | | |
| Position 05: | A | E | G | I | M | R | V | C | | |
| Position 06: | C | E | K | L | N | R | V | | | |
| Position 07: | A | C | E | I | L | S | T | V | W | |
| Position 08: | H | K | L | M | S | T | W | Y | | |
| Position 09: | A | I | L | M | Q | S | R | | | |
| Position 10: | A | I | M | S | W | Y | | | | |
| Position 11: | C | I | K | L | S | V | | | | |
| Position 12: | C | E | K | L | P | Q | R | T | | |
| Position 13: | A | D | H | N | W | | | | | |
| Position 14: | A | C | G | I | L | S | T | V | M | |
| Position 15: | A | E | G | I | K | L | M | R | V | |
| Position 16: | G | H | L | R | S | T | V | C | | |
| Position 17: | A | I | L | V | | | | | | |
| Position 18: | A | C | D | E | G | H | I | K | M | S |
| Position 19: | D | E | G | L | N | V | T | | | |

15. The polypeptide of any one of claims 1-14, wherein the polypeptide comprises an amino acid sequence at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, at least 98%, or 100% identical to the amino acid sequence set forth in SEQ ID NOS: 76, 168, 90, 181, 91, 182, 94 or 246 wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$) or SEQ ID NOS: 92, 93, 183, or 184 wherein the polypeptide binds to IL-4 receptor $\alpha\gamma_c$heterodimer (IL-4R$\alpha\gamma_c$).

16. A non-naturally occurring polypeptide comprising an amino acid sequence at least 90%, at least 95%, at least 98% or 100% identical to the amino acid sequence set forth in SEQ ID NO: 90 wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$).

17. A non-naturally occurring polypeptide comprising an amino acid sequence at least 90%, at least 95%, at least 98% or 100% identical to the amino acid sequence set forth in SEQ ID NO: 181 wherein the polypeptide binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-

2Rβγc).

18. The polypeptide of any one of claims 1-5, wherein the polypeptide comprises an amino acid sequence at least 60%, at least 65%, at least 70%, at least 75%, at least 80%, at least 85%, at least 90%, at least 95%, or at least 98%, identical to the amino acid sequence set forth in SEQ ID NO:181, and wherein one of the following mutations are present

D56C;
K58C;
D59C;
R66C;
T77C;
E85C;
R50C;
E53C;
E62C;
E69C;
R73C; or
E82C.

19. The polypeptide of any one of claims 1-18, wherein the polypeptide is linked to a stabilization compound, including but not limited to a polyethylene glycol ("PEG") containing moiety.

20. The polypeptide of claim 19, wherein the polypeptide comprises a cysteine residue and one or more polyethylene glycol chains is linked to the cysteine residue via a maleimide group.

21. The polypeptide of any one of claims 1-7, or 9-19 wherein the polypeptide binds to the human IL-2 receptor βγc heterodimer (IL-2Rβγc) and wherein the polypeptide's affinity for the human IL-2 receptor βγc heterodimer is greater than that of native IL-2.

22. The polypeptide of claim 21, wherein the polypeptide stimulates STAT5 phosphorylation in cells expressing the human IL-2 receptor βγc heterodimer (IL-2Rβγc) with potency greater than native IL-2.

23. The polypeptide of claim 1, comprising the amino acid sequence set forth in SEQ ID NO:190, 191, 195, 197, 198, or SEQ ID NO:200.

24. The polypeptide of any one of claims 1-22, wherein the polypeptide includes at least one disulfide bond.

25. The polypeptide of claim 1 comprising the amino acid sequence set forth in SEQ ID NO:195 wherein a polyethylene glycol containing moiety is linked to the cysteine reside at position 62 of SEQ ID NO:195.

26. The polypeptide of any one of claims 1-25, wherein the polypeptide further comprises a targeting domain.

27. The polypeptide of claim 26, wherein the targeting domain is a translational fusion with the polypeptide.

28. The polypeptide of claim 26 or claim 27, wherein the targeting domain binds to a cell surface protein present on the surface of a tumor cell, a tumor vascular component cell, or a tumor microenvironment cell .

29. The polypeptide of claim 26 or claim 27, wherein the targeting domain binds to a tumor cell, tumor vascular component cell, or tumor microenvironment cell surface marker wherein the cell surface marker is EGFR, EGFRvIII, Her2, HER3, EpCAM, MSLN, MUC16, PSMA, TROP2, ROR1, RON, PD-L1, CD47, CTLA-4,CD5, CD19, CD20, CD25, CD37, CD30, CD33, CD40, CD45, CAMPATH-1, BCMA, CS-1, PD-L1, B7-H3, B7-DC, HLD-DR, carcinoembryonic antigen (CEA), TAG-72, EpCAM, MUC1, folate-binding protein, A33, G250, prostate-specific membrane antigen (PSMA), ferritin, GD2, GD3, GM2, $Le^y$, CA-125, CA19-9, epidermal growth factor, p185HER2, IL-2 receptor, EGFRvIII (de2-7 EGFR), fibroblast activation protein, tenascin, a metalloproteinase, endosialin, vascular endothelial growth factor, avB3, WT1, LMP2, HPV E6, HPV E7, Her-2/neu, MAGE A3, p53 nonmutant, NY-ESO-1, MelanA/MART1, Ras mutant, gp100, p53 mutant, PR1, bcr-abl, tyronsinase, survivin, PSA, hTERT, a Sarcoma translocation breakpoint protein, EphA2, PAP, ML-IAP, AFP, ERG, NA17, PAX3, ALK, androgen receptor, cyclin B 1, polysialic acid, MYCN, RhoC, TRP-2, fucosyl GM1, mesothelin (MSLN), PSCA, MAGE Al, sLe(animal), CYP1B1,

PLAV1, GM3, BORIS, Tn, GloboH, ETV6-AML, NY-BR-1, RGS5, SART3, STn, Carbonic anhydrase IX, PAX5, OY-TESL Sperm protein 17, LCK, HMWMAA, AKAP-4, SSX2, XAGE 1, Legumain, Tie 3, VEGFR2, MAD-CT-1, PDGFR-B, MAD-CT-2, ROR2, TRAIL1, MUC16, MAGE A4, MAGE C2, GAGE, EGFR, CMET, HER3, MUC15, CA6, NAPI2B, TROP2, CLDN6, CLDN16, CLDN18.2, CLorf186, RON, LY6E, FRA, DLL3, PTK7, STRA6, TMPRSS3, TMPRSS4, TMEM238, UPK1B, VTCN1, LIV1, ROR1, Fos-related antigen 1, or one of the markers listed below:

(1) BMPR1B (bone morphogenetic protein receptor-type IB, Genbank accession no. NM.sub.--001203);

(2) E16 (LAT1, SLC7A5, Genbank accession no. NM.sub.--003486);

(3) STEAP1 (six transmembrane epithelial antigen of prostate, Genbank accession no. NM.sub.--012449);

(4) 0772P (CA125, MUC16, Genbank accession no. AF361486);

(5) MPF (MPF, MSLN, SMR, megakaryocyte potentiating factor, mesothelin, Genbank accession no. NM.sub.--005823);

(6) Napi3b (NAPI-3B, NPTIIb, SLC34A2, solute carrier family 34 (sodium phosphate), member 2, type II sodium-dependent phosphate transporter 3b, Genbank accession no. NM.sub.--006424);

(7) Sema 5b (FLJ10372, KIAA1445, Mm. 42015, SEMA5B, SEMAG, Semaphorin 5b Hlog, sema domain, seven thrombospondin repeats (type 1 and type 1-like), transmembrane domain (TM) and short cytoplasmic domain, (semaphorin) 5B, Genbank accession no. AB040878);

(8) PSCA hlg (2700050C12Rik, C530008O16Rik, RIKEN cDNA 2700050C12, RIKEN cDNA 2700050C12 gene, Genbank accession no. AY358628);

(9) ETBR (Endothelin type B receptor, Genbank accession no. AY275463);

(10) MSG783 (RNF124, hypothetical protein FLJ20315, Genbank accession no. NM.sub.--017763);

(11) STEAP2 (HGNC.sub.--8639, IPCA-1, PCANAP1, STAMP1, STEAP2, STMP, prostate cancer associated gene 1, prostate cancer associated protein 1, six transmembrane epithelial antigen of prostate 2, six transmembrane prostate protein, Genbank accession no. AF455138);

(12) TrpM4 (BR22450, FLJ20041, TRPM4, TRPM4B, transient receptor potential cation channel, subfamily M, member 4, Genbank accession no. NM.sub.--

017636);

(13) CRIPTO (CR, CR1, CRGF, CRIPTO, TDGF1, teratocarcinoma-derived growth factor, Genbank accession no. NP.sub.--003203 or NM.sub.--003212);

(14) CD21 (CR2 (Complement receptor 2) or C3DR(C3d/Epstein Barr virus receptor) or Hs. 73792, Genbank accession no. M26004);

(15) CD79b (IGb (immunoglobulin-associated beta), B29, Genbank accession no. NM.sub.--000626);

(16) FcRH2 (IFGP4, IRTA4, SPAP1A (SH2 domain containing phosphatase anchor protein 1a), SPAP1B, SPAP1C, Genbank accession no. NM.sub.--030764);

(17) HER2 (Genbank accession no. M11730);

(18) NCA (Genbank accession no. M18728);

(19) MDP (Genbank accession no. BC017023);

(20) IL20R.alpha. (Genbank accession no. AF184971);

(21) Brevican (Genbank accession no. AF229053);

(22) Ephb2R (Genbank accession no. NM.sub.--004442);

(23) ASLG659 (Genbank accession no. AX092328);

(24) PSCA (Genbank accession no. AJ297436);

(25) GEDA (Genbank accession no. AY260763);

(26) BAFF-R (Genbank accession no. NP.sub.--443177.1);

(27) CD22 (Genbank accession no. NP-001762.1);

(28) CD79a (CD79A, CD79.alpha., immunoglobulin-associated alpha, a B cell-specific protein that covalently interacts with Ig beta (CD79B) and forms a complex on the surface with Ig M molecules, transduces a signal involved in B-cell differentiation, Genbank accession No. NP.sub.--001774.1);

(29) CXCR5 (Burkitt's lymphoma receptor 1, a G protein-coupled receptor that is activated by the CXCL13 chemokine, functions in lymphocyte migration and humoral defense, plays a role in HIV-2 infection and perhaps development of AIDS, lymphoma, myeloma, and leukemia, Genbank accession No. NP.sub.--001707.1);

(30) HLA-DOB (Beta subunit of MHC class II molecule (Ia antigen) that binds peptides and presents them to CD4+ T lymphocytes, Genbank accession No. NP.sub.--002111.1);

(31) P2X5 (Purinergic receptor P2X ligand-gated ion channel 5, an ion channel gated by extracellular ATP, may be involved in synaptic transmission and neurogenesis,

deficiency may contribute to the pathophysiology of idiopathic detrusor instability, Genbank accession No. NP.sub.--002552.2);

(32) CD72 (B-cell differentiation antigen CD72, Lyb-2, Genbank accession No. NP.sub.--001773.1);

(33) LY64 (Lymphocyte antigen 64 (RP105), type I membrane protein of the leucine rich repeat (LRR) family, regulates B-cell activation and apoptosis, loss of function is associated with increased disease activity in patients with systemic lupus erythematosis, Genbank accession No. NP.sub.--005573.1);

(34) FCRH1 (Fc receptor-like protein 1, a putative receptor for the immunoglobulin Fc domain that contains C2 type Ig-like and ITAM domains, may have a role in B-lymphocyte differentiation, Genbank accession No. NP.sub.--443170.1); or

(35) IRTA2 (Immunoglobulin superfamily receptor translocation associated 2, a putative immunoreceptor with possible roles in B cell development and lymphomagenesis; deregulation of the gene by translocation occurs in some B cell malignancies, Genbank accession No. NP.sub.--112571.1).

30. The polypeptide of claim 26 or 27, wherein the targeting domain binds to an immune cell surface marker.

31. A recombinant nucleic acid encoding the polypeptide of any one of claims 1-30.

32. A recombinant host cell comprising the nucleic acid of claim 31.

33. A pharmaceutical composition comprising a polypeptide of any one of claims 1-7 or 9-30 that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$) or a recombinant nucleic acid of claim 31 that encodes a polypeptide that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), and a pharmaceutically acceptable carrier.

34. Use of a polypeptide of any one of claims 1-7 or 9-30 that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), a recombinant nucleic acid of claim 31 that encodes a polypeptide that binds to IL-2 receptor $\beta\gamma_c$ heterodimer (IL-2R$\beta\gamma_c$), or the pharmaceutical composition of claim 33 in the manufacture of a medicament for treating cancer that expresses IL-2 receptor $\beta\gamma$c heterodimer (IL-2R$\beta\gamma$c) in a subject having said cancer.