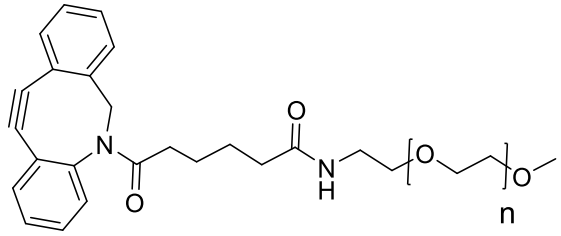
BRIEF SUMMARY

The present disclosure provides herein interleukin 2 (IL-2) polypeptides having at least one or more amino acid substitutions that bias binding to the IL-2 receptor $\beta\gamma_c$ dimer (IL-2R$\beta\gamma_c$) over the IL-2 receptor $\alpha\beta\gamma_c$ trimer (IL-2R$\alpha\beta\gamma_c$), a non-natural amino acid at or near the N-terminus of the IL-2 polypeptide conjugated to a nonpeptidic, water-soluble polymer, and an amino acid sequence with at least 80% identity otherwise to the amino acid sequence wild-type or native human IL-2. These IL-2 conjugates have a binding affinity to the IL-2R$\beta\gamma_c$ that is substantially similar to that of native IL-2 and has reduced or no detectable binding affinity for the IL-2Rα compared to native IL-2 as determined by Surface Plasmon Resonance (SPR) assay. Thus, these IL-2 conjugates have reduced toxicity compared to compared to commercially available IL-2 polypeptides and a plasma half-life of at least five times that of commercially available IL-2 polypeptides.

The present disclosure provides an interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80%, 85%, 90%, 95%, 98%, or 99% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising: (i) one or more amino acid substitutions that reduce(s) affinity of the IL-2 polypeptide for the human IL-2 receptor $\alpha\beta\gamma_c$ trimer (IL-2R$\alpha\beta\gamma_c$) relative to wild-type human IL-2; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide; wherein the IL-2 polypeptide has substantially similar binding affinity for the human IL-2 receptor $\beta\gamma_c$ dimer (IL-2R$\beta\gamma_c$) relative to wild-type human IL-2.

In one aspect, the present disclosure provides an interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising: (i) one or more amino acid substitutions at amino acid positions R37 and F41 wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide or a position within the first 10 amino acids comprising the IL-2 polypeptide with a non-natural amino acid comprising an azide group,

wherein the azide group is conjugated to an alkyne group of a nonpeptidic, water-soluble polymer having the formula:



wherein n is an integer from 1 to 5,000; wherein the IL-2 polypeptide has substantially similar binding affinity for the human IL-2 receptor βγc dimer (IL-2Rβγc) relative to wild-type human IL-2.

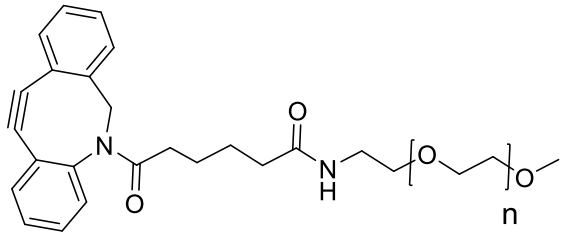
In further examples, the IL-2 polypeptide amino acid sequence comprises at least 80%, 85%, 90%, 95%, 98%, or 99% identity with amino acid sequence set forth in SEQ ID: 2 with the proviso that the IL-2 polypeptide comprises at least amino acids E15, H16, L19, D20, D84, N88, V91, Q126, T123, and I129, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In further examples, the IL-2 polypeptide amino acid sequence comprises at least 80% identity with amino acid sequence set forth in SEQ ID: 2 with the proviso that the IL-2 polypeptide comprises at least amino acids E15, H16, L19, D20, D84, N88, V91, Q126, T123, and I129, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53. In further examples, the IL-2 polypeptide amino acid sequence

wherein RG is a reactive group that is capable of forming a covalent linkage with the functional group of a non-natural amino acid; linker is a covalent bond or a substituted or non-substituted C_{1-20} alkyl; POLY is a nonpeptidic, water-soluble polymer; and x is an alcohol or methyl group at the terminus of the POLY.

In a further example, the reactive group of the nonpeptidic, water-soluble polymer comprises an alkyne and the functional group of the non-natural amino acid comprises an azide or the reactive group of the nonpeptidic, water-soluble polymer comprises an azide and the functional group of the non-natural amino acid comprises an alkyne.

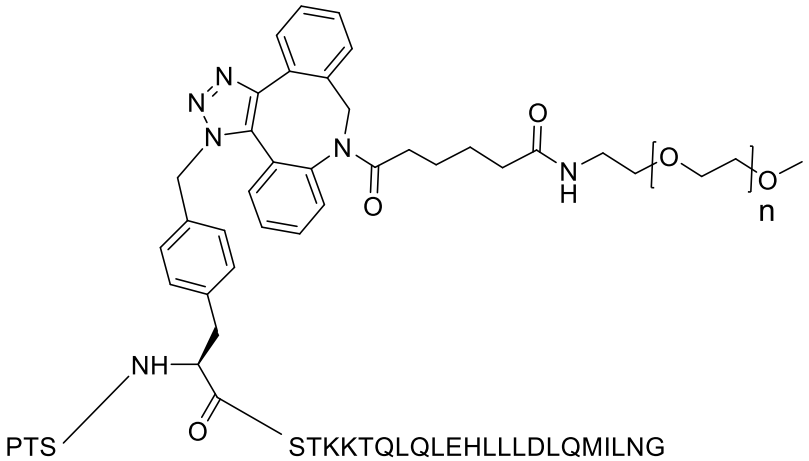
In a further example, the nonpeptidic, water-soluble polymer conjugated to the reactive group has the formula:



wherein n is about 681.

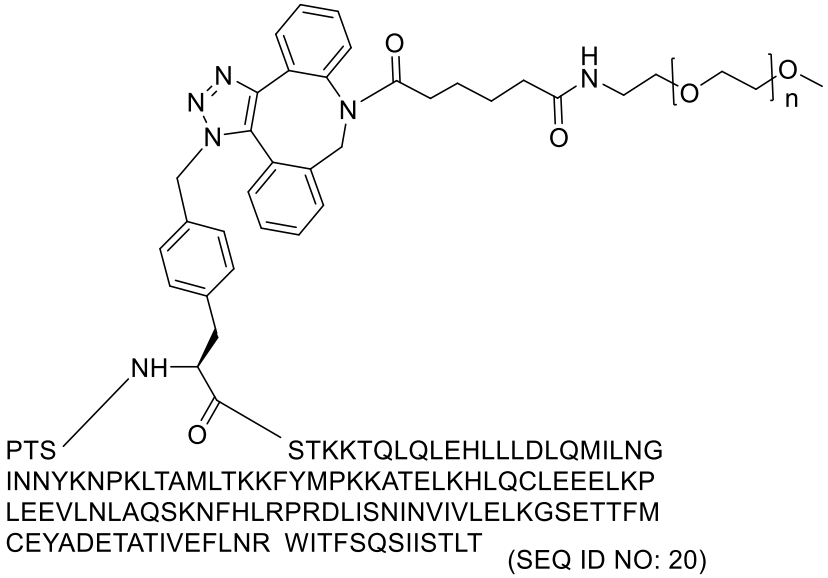
In a further example, the IL-2 conjugate comprises the amino acid sequence set forth in SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 16, SEQ ID NO: 17, SEQ ID NO: 18, SEQ ID NO: 19, SEQ ID NO: 20, or SEQ ID NO: 21.

The present disclosure provides an interleukin 2 (IL-2) conjugate comprising the formula:



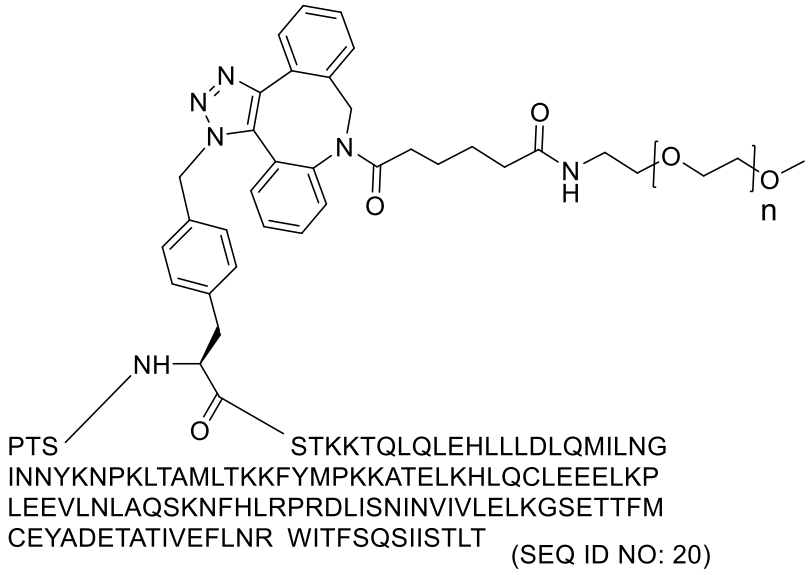
(SEQ ID NO: 20)

wherein n is about 681, or a regioisomer thereof comprising the formula

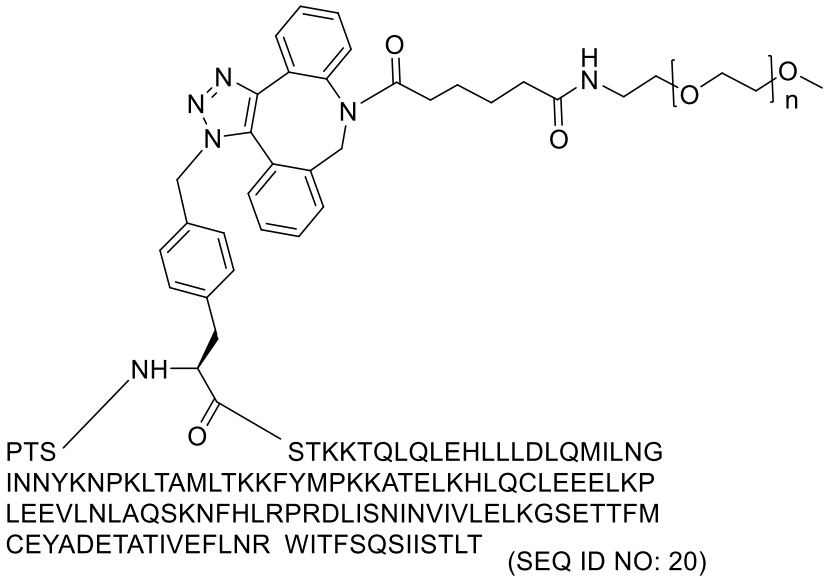


wherein n is about 681.

In a further aspect, the present disclosure provides an interleukin 2 (IL-2) conjugate comprising the formula



wherein n is an integer from 1 to 5,000, or a regioisomer thereof comprising the formula



wherein n is an integer from 1 to 5,000.

The present disclosure provides a composition comprising: the IL-2 conjugate as described herein and a pharmaceutically acceptable carrier or excipient.

In a further example, the present disclosure provides a method for treating a proliferative disease or cancer in an individual, comprising: administering a therapeutically effective amount of any example of IL-2 conjugate disclosed herein or composition thereof to an individual in need thereof to treat the proliferative disease or cancer in the individual.

In a further aspect, the present disclosure provides the IL-2 conjugate as defined herein for use in therapy.

In a further aspect, the present disclosure provides the IL-2 conjugate as defined herein for use in combination with a therapeutic agent selected from checkpoint inhibitor and chemotherapy agent for use in therapy.

In a further example, the present disclosure provides a combination therapy for treating a proliferative disease or cancer in an individual, comprising: administering a therapeutically effective amount of any example of IL-2 conjugate disclosed herein or composition thereof to an individual in need thereof, and administering a therapeutically effective amount of a therapeutic agent to the individual, to treat the proliferative disease or cancer in the individual. In a further example, the therapeutic agent is an anti-PD1 antibody or anti-PDL1 antibody. In a further example, the IL-2 conjugate or composition is administered before the therapeutic agent is administered. In a further example, the IL-2 conjugate or composition is administered after the therapeutic agent is administered. In a further example,

the IL-2 conjugate or composition is administered concurrently with the therapeutic agent.

The present disclosure further provides for the use of the IL-2 conjugate of any example of IL-2 conjugate disclosed herein or composition thereof for the treatment of a

proliferative disease or cancer. In a further example, is provided the use of the IL-2 conjugate of any example of IL-2 conjugate disclosed herein or composition thereof and a therapeutic agent for the treatment of a proliferative disease or cancer. In a further example, the therapeutic agent is an anti-PD1 antibody or anti-PDL1 antibody.

The present disclosure further provides for the use of the IL-2 conjugate of any example of IL-2 conjugate disclosed herein or composition thereof for the manufacture of a medicament for the treatment of a proliferative disease or cancer.

In a further aspect, the present disclosure provides use of the IL-2 conjugate as defined herein in the manufacture of a medicament for the treatment of a proliferative disease or cancer.

In a further aspect, the present disclosure provides use of the IL-2 conjugate as defined herein in the manufacture of a medicament for use with a therapeutic agent selected from checkpoint inhibitor and chemotherapy agent in a combination therapy for treatment of a proliferative disease or cancer.

The present disclosure further provides an IL-2 conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising: (i) one or more amino acid substitutions selected from the group consisting of K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, M103, C104, and Y106; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In particular examples, the IL-2 polypeptide comprises an amino acid sequence with at least 85% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprises: (i) one or more amino acid substitutions selected from the group consisting of K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, M103, C104, and Y106; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In particular examples, the IL-2 polypeptide comprises an amino acid sequence with at least 90% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprises: (i) one or more amino acid substitutions selected from the group consisting of K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, M103, C104, and Y106; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-

terminus of the IL-2 polypeptide, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In particular examples, the IL-2 polypeptide comprises an amino acid sequence with at least 95% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprises: (i) one or more amino acid substitutions selected from the group consisting of K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, M103, C104, and Y106; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In a further aspect, the present disclosure provides an interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80%, 85%, 90%, 95%, 98%, or 99% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising: (i) one or more amino acid substitutions selected from the group consisting of K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, M103, C104, and Y106; and (ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In a further example, the one or more amino acid substitutions in the IL-2 polypeptide are at positions R37 and F41. In a further example, the amino acid substitutions in the IL-2 polypeptide are R37A and F41K. In a further example, the IL-2 polypeptide further includes a substitution of the cysteine residue at position 124 with an amino acid selected from the group consisting of A and S, wherein the amino acid position corresponds to the position of the amino acid in the amino acid sequence set forth in SEQ ID NO: 53. In a further example, the IL-2 polypeptide further includes an N-terminal alanine residue.

In a further example, the non-natural amino acid is substituted for an amino acid corresponding to a position within the first 10 amino acids of the amino acid sequence as set forth in SEQ ID NO: 2 or inserted within said sequence. In a further example, the non-natural amino acid is substituted for an amino acid corresponding to a position within the sequence from P1 to Q10 of the amino acid sequence as set forth in SEQ ID NO: 2 or inserted within said sequence between P1 through Q10. In a further example, the non-natural amino acid is

substituted for an amino acid at position P1, T2, S3, S4, S5, T6, K7, K8, or T9 or linked to the N-terminal amino acid by an amide linkage. In a further example, the non-natural amino acid is located at the amino acid position corresponding to position 4 of the amino acid sequence set forth in SEQ ID NO: 2.

In a further example, the non-natural amino acid comprises a functional group and the nonpeptidic, water-soluble polymer is linked to a reactive group that is capable of reacting with the functional group to form a covalent linkage. In a further example, the non-natural amino acid is selected from the group consisting of p-azidomethyl-L-phenylalanine, p-azido-L-phenylalanine, p-acetyl-L-phenylalanine, N6-azidoethoxy-L-lysine, N6-propargylethoxy- L-lysine (PraK), BCN-L-lysine, norbornene lysine, TCO-lysine,

for the treatment of a proliferative disease or cancer. In a further example, the therapeutic agent is an anti-PD1 antibody or anti-PDL1 antibody.

The present disclosure further provides for the use of the IL-2 conjugate of any example of IL-2 conjugate disclosed herein or composition thereof for the manufacture of a medicament for the treatment of a proliferative disease or cancer.

In a further aspect, the present disclosure provides an IL-2 conjugate comprising the amino acid sequence set forth in SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 16, SEQ ID NO: 17, SEQ ID NO: 18, SEQ ID NO: 19, SEQ ID NO: 20, or SEQ ID NO: 21. In a further example, the present disclosure provides a pharmaceutical composition comprising the IL-2 conjugate and a pharmaceutically acceptable carrier or excipient.

In a further aspect, the present disclosure further provides an IL-2 moiety comprising an amino acid sequence set forth in SEQ ID NO: 42, SEQ ID NO: 43, SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 50, SEQ ID NO: 51, or SEQ ID NO: 52.

The present disclosure further provides an interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80%, 85%, 90%, 95%, 98%, or 99% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising: a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, with the proviso that the IL-2 polypeptide comprises at least amino acids E15, H16, L19, D20, K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, D84, N88, V91, M103, C104, Y106, Q126, T123, and I129, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

In a further example, the IL-2 polypeptide further includes a substitution of the cysteine residue at position 124 with an amino acid selected from the group consisting of A and S.

In a further example, the IL-2 polypeptide further includes an N-terminal alanine residue.

In a further example, the non-natural amino acid is substituted for an amino acid corresponding to a position within the first 10 amino acids of the amino acid sequence as set

forth in SEQ ID NO: 2 or inserted within said sequence. In a further example, the non-natural amino acid is substituted for an amino acid corresponding to a position within the sequence from P1 to Q10 of the amino acid sequence as set forth in SEQ ID NO: 2 or inserted within said sequence between P1 through Q10. In a further example, the non-natural amino acid is substituted for an amino acid at position P1, T2, S3, S4, S5, T6, K7, K8, or T9 or linked to the N-terminal amino acid by an amide linkage. In a further example, the non-natural amino acid is located at the amino acid position corresponding to position 4 of the amino acid sequence set forth in SEQ ID NO: 2.

In a further aspect, the present disclosure provides an interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80%, 85%, 90%, 95%, 98%, or 99% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising: a substitution of an amino acid at position P1, T2, S3, S4, S5, T6, K7, K8, or T9 or linked to the N-terminal amino acid of the IL-2 polypeptide with a non-natural amino acid conjugated by an amide bond to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, with the proviso that the IL-2 polypeptide comprises at least amino acids E15, H16, L19, D20, K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, D84, N88, V91, M103, C104, Y106, Q126, T123, and I129, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53 according to numbering scheme B.

In a further example, the non-natural amino acid comprises a functional group and the water-soluble polymer is linked to a reactive group that is capable of reacting with the functional group to form a covalent linkage.

In a further example, the non-natural amino acid is selected from the group consisting of p-azidomethyl-L-phenylalanine, p-azido-L-phenylalanine, p-acetyl-L-phenylalanine, N6-azidoethoxy-L-lysine, N6-propargylethoxy- L-lysine (PraK), BCN-L-lysine, norbornene lysine, TCO-lysine, methyltetrazine lysine, allyloxycarbonyllysine, 2-amino-8-oxononanoic acid, 2-amino-8-oxooctanoic acid, O-methyl-L-tyrosine, L-3-(2-naphthyl)alanine, 3-methyl-phenylalanine, O-4-allyl-L-tyrosine, 4-propyl-L-tyrosine, tri-O-acetyl-GlcNAc-serine, L-Dopa, fluorinated phenylalanine, isopropyl-L-phenylalanine, p-benzoyl-L-phenylalanine, L-phosphoserine, phosphonoserine, phosphonotyrosine, p-iodo-phenylalanine, p-bromophenylalanine, p-amino-L-phenylalanine, isopropyl-L-phenylalanine, p-propargyloxy-phenylalanine, 2-amino-3-((2-((3- (benzyloxy)-3-oxopropyl)amino)ethyl)selanyl)propanoic acid, 2-amino-3- (phenylselanyl)propanoic, selenocysteine, m-acetylphenylalanine, 2-amino-8-

oxononanoic acid, and p-propargyloxyphenylalanine. In a further example, the non-natural amino acid is p-azidomethyl-L-phenylalanine.

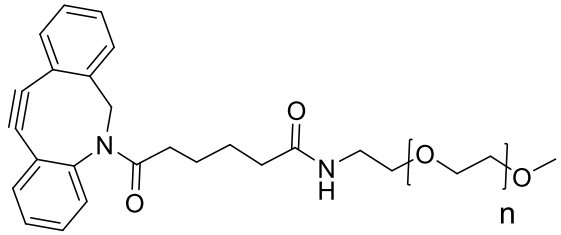
In a further example, the nonpeptidic, water-soluble polymer has an average molecular weight between 1 kDa and 100 kDa. I n a further example, the nonpeptidic, water-soluble polymer has an average molecular weight of about 30 kDa. In a further example, The IL-2 conjugate of claim 65, wherein the nonpeptidic, water-soluble polymer is polyethylene glycol (PEG), poly(propylene glycol) (PPG), copolymers of ethylene glycol and propylene glycol, poly(oxyethylated polyol), poly(olefinic alcohol), poly(vinylpyrrolidone), poly(hydroxyalkylmethacrylamide), poly(hydroxyalkylmethacrylate), poly(saccharides), poly(a-hydroxy acid), poly(vinyl alcohol), polyphosphazene, polyoxazolines (POZ), poly(N-acryloylmorpholine), or a combination thereof. In a further example, the nonpeptidic, water-soluble polymer is a linear or branched PEG.

WHAT IS CLAIMED:

1. An interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising:

(i) one or more amino acid substitutions at amino acid positions R37 and F41 wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53; and

(ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide or a position within the first 10 amino acids comprising the IL-2 polypeptide with a non-natural amino acid comprising an azide group, wherein the azide group is conjugated to an alkyne group of a nonpeptidic, water-soluble polymer having the formula:



wherein n is an integer from 1 to 5,000;

wherein the IL-2 polypeptide has substantially similar binding affinity for the human IL-2 receptor $\beta\gamma_c$ dimer (IL-2R$\beta\gamma_c$) relative to wild-type human IL-2.

2. The IL-2 conjugate of claim 1, wherein the IL-2 polypeptide conjugate has no detectable binding to the human IL-2 receptor α monomer (IL-2Rα) as determined by a Surface Plasmon Resonance assay.

3. The IL-2 conjugate of claim 1, wherein n is 681.

4. The IL-2 conjugate of claim 1, wherein the amino acid substitutions in the IL-2 polypeptide are R37A and F41K.

5. The IL-2 conjugate of claim 1, wherein the IL-2 polypeptide further includes an amino acid substitution of the cysteine residue at position 124 with an amino acid selected from the group consisting of A and S, wherein the amino acid position corresponds to the position of the amino acid in the amino acid sequence set forth in SEQ ID NO: 53.

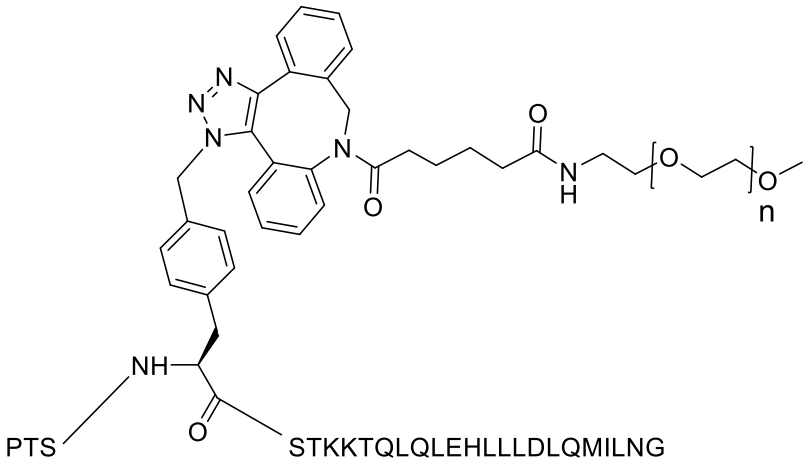
6. The IL-2 conjugate of claim 1, wherein the IL-2 polypeptide further includes an N-terminal alanine residue or N-terminal methionine residue.

7. The IL-2 conjugate of claim 1, wherein the non-natural amino acid is selected from the group consisting of p-azidomethyl-L-phenylalanine, p-azido-L-phenylalanine, p-acetyl-L-phenylalanine, and N6-azidoethoxy-L-lysine.

8. The IL-2 conjugate of claim 1, wherein the non-natural amino acid is p-azidomethyl-L-phenylalanine.

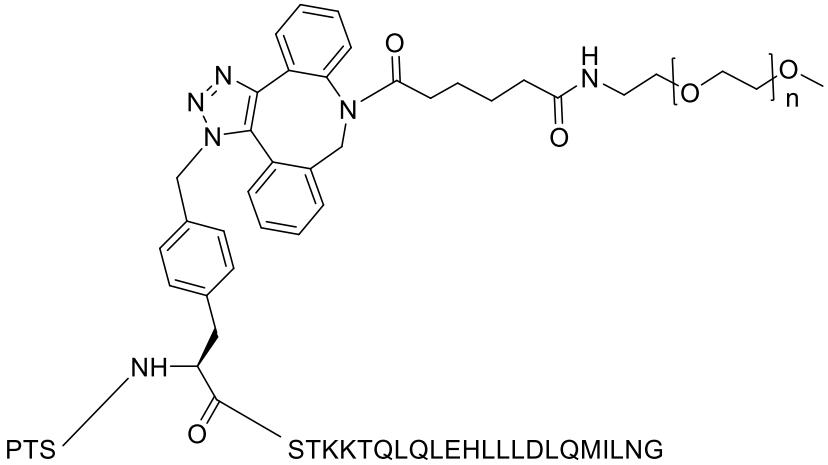
9. The IL-2 conjugate of claim 1, wherein the IL-2 conjugate comprises the amino acid sequence set forth in SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 16, SEQ ID NO: 17, SEQ ID NO: 18, SEQ ID NO: 19, SEQ ID NO: 20, or SEQ ID NO: 21.

10. An interleukin 2 (IL-2) conjugate comprising the formula



(SEQ ID NO: 20)

wherein n is an integer from 1 to 5,000, or a regioisomer thereof comprising the formula



(SEQ ID NO: 20)

wherein n is an integer from 1 to 5,000.

11. The interleukin 2 (IL-2) conjugate of claim 10, wherein the integer is 681.

12. The IL-2 conjugate of any one of claims 1-11 for use in therapy.

13. The IL-2 conjugate of any one of claims 1-11 for use in combination with a therapeutic agent selected from checkpoint inhibitor and chemotherapy agent for use in therapy.

14. Use of the IL-2 conjugate of any one of claims 1-11 in the manufacture of a medicament for the treatment of a proliferative disease or cancer.

15. Use of the IL-2 conjugate of any one of claims 1-11 in the manufacture of a medicament for use with a therapeutic agent selected from checkpoint inhibitor and chemotherapy agent in a combination therapy for treatment of a proliferative disease or cancer.

16. The IL-2 conjugate for use of claim 13 or the use of claim 15, wherein the therapeutic agent is an anti-PD1 antibody or anti-PDL1 antibody.

17. An interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80%, 85%, 90%, 95%, 98%, or 99% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising:

(i) one or more amino acid substitutions selected from the group consisting of K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, M103, C104, and Y106; and

(ii) a substitution of an amino acid at or near the N-terminus of the IL-2 polypeptide with a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53.

18. The IL-2 conjugate of claim 17 for use in therapy.

19. The IL-2 conjugate of claim 17 and a therapeutic agent selected from checkpoint inhibitor and chemotherapy agent for use in therapy.

20. Use of the IL-2 conjugate of claim 17 in the manufacture of a medicament for the treatment of a proliferative disease or cancer.

21. Use of the IL-2 conjugate of claim 17 in the manufacture of a medicament for use with a therapeutic agent selected from checkpoint inhibitor and chemotherapy agent in a combination therapy for treatment of a proliferative disease or cancer.

22. The IL-2 conjugate for use of claim 19 or the use of claim 21, wherein the therapeutic agent is an anti-PD1 antibody or anti-PDL1 antibody.

23. An IL-2 conjugate comprising the amino acid sequence set forth in SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9, SEQ ID NO: 10, SEQ ID NO: 11, SEQ ID NO: 12, SEQ ID NO: 13, SEQ ID NO: 14, SEQ ID NO: 15, SEQ ID NO: 16, SEQ ID NO: 17, SEQ ID NO: 18, SEQ ID NO: 19, SEQ ID NO: 20, or SEQ ID NO: 21.

24. An IL-2 moiety comprising an amino acid sequence set forth in SEQ ID NO: 42, SEQ ID NO: 43, SEQ ID NO: 44, SEQ ID NO: 45, SEQ ID NO: 46, SEQ ID NO: 47, SEQ ID NO: 48, SEQ ID NO: 49, SEQ ID NO: 50, SEQ ID NO: 51, or SEQ ID NO: 52.

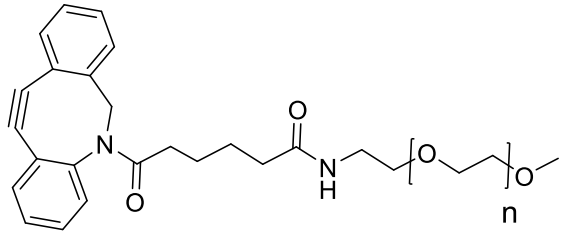
25. An interleukin 2 (IL-2) conjugate comprising an IL-2 polypeptide comprising an amino acid sequence with at least 80%, 85%, 90%, 95%, 98%, or 99% identity to the amino acid sequence set forth in SEQ ID NO: 53 and further comprising:

a substitution of an amino acid at position P1, T2, S3, S4, S5, T6, K7, K8, or T9 or linked to the N-terminal amino acid of the IL-2 polypeptide with a non-natural amino acid conjugated by an amide bond to a nonpeptidic, water-soluble polymer or an insertion of a non-natural amino acid conjugated to a nonpeptidic, water-soluble polymer at or near the N-terminus of the IL-2 polypeptide, with the proviso that the IL-2 polypeptide comprises at least amino acids E15, H16, L19, D20, K34, T36, R37, T40, F41, K42, F43, Y44, E60, E61, K63, P64, E67, L71, D84, N88, V91, M103, C104, Y106, Q126, T123, and I129, wherein the amino acid positions correspond to the positions set forth in the amino acid sequence of SEQ ID NO: 53 according to numbering scheme B.

26. The IL-2 polypeptide conjugate of claim 25, wherein the non-natural amino acid is selected from the group consisting of p-azidomethyl-L-phenylalanine, p-azido-L-phenylalanine, p-acetyl-L-phenylalanine, N6-azidoethoxy-L-lysine, N6-propargylethoxy- L-lysine (PraK), BCN-L-lysine, norbornene lysine, TCO-lysine, methyltetrazine lysine, allyloxycarbonyllysine, 2-amino-8-oxononanoic acid, 2-amino-8-oxooctanoic acid, O-methyl-L-tyrosine, L-3-(2-naphthyl)alanine, 3-methyl-phenylalanine, O-4-allyl-L-tyrosine, 4-propyl-L-tyrosine, tri-O-acetyl-GlcNAc-serine, L-Dopa, fluorinated phenylalanine, isopropyl-L-phenylalanine, p-benzoyl-L-phenylalanine, L-phosphoserine, phosphonoserine, phosphonotyrosine, p-iodo-phenylalanine, p-bromophenylalanine, p-amino-L-phenylalanine, isopropyl-L-phenylalanine, p-propargyloxy-phenylalanine, 2-amino-3-((2-((3- (benzyloxy)-3-oxopropyl)amino)ethyl)selanyl)propanoic acid, 2-amino-3- (phenylselanyl)propanoic, selenocysteine, m-acetylphenylalanine, and p-propargyloxyphenylalanine.

27. The IL-2 conjugate of claim 25, wherein the nonpeptidic, water-soluble polymer is polyethylene glycol (PEG), poly(propylene glycol) (PPG), copolymers of ethylene glycol and propylene glycol, poly(oxyethylated polyol), poly(olefinic alcohol), poly(vinylpyrrolidone), poly(hydroxyalkylmethacrylamide), poly(hydroxyalkylmethacrylate), poly(saccharides), poly(a-hydroxy acid), poly(vinyl alcohol), polyphosphazene, polyoxazolines (POZ), poly(N-acryloylmorpholine), or a combination thereof.

28. The IL-2 conjugate of claim 25, wherein the nonpeptidic, water-soluble polymer, which comprises the alkyne, has the formula:



wherein n is an integer from 1 to 5,000.