

In some embodiments, the IL-2 muteins possess decreased binding affinity to the extracellular domain of hCD132 while retaining significant binding to the hCD25/hCD122 receptor complex and/or activation of the hCD25/hCD122/hCD132 receptor complex.

5 In some embodiments, the IL-2 muteins possess decreased binding affinity to CD132 while retaining substantial binding affinity for hCD25.

In one aspect, the present disclosure provides hIL-2 muteins exhibiting significant or enhanced binding affinity for hCD25 and reduced binding affinity for the extracellular domain of hCD132 receptor as compared to wild type human IL-2 (hIL-2). In some embodiments, the IL-2 muteins comprise one or more amino acid substitutions that decrease
10 CD132 receptor binding affinity selected from amino acid positions 18, 22, and 126, numbered in accordance with mature wild type hIL-2.

In one aspect, the present disclosure provides polypeptide having decreased binding affinity, compared to a wild type hIL2, to the extracellular domain of hCD132, the polypeptide comprising an amino acid sequence of the formula:

15 (AA1)_a-(AA2)_b-(AA3)_c-(AA4)_d-(AA5)_e-(AA6)_f-(AA7)_g-(AA8)_h-(AA9)_i-
T10-Q11-L12-Q13-L14-E15-H16-L17-(AA18)-L19-D20-L21-(AA22)-
M23-I24-L25-N26-G27-I28-N29-N30-Y31-K32-N33-P34-(AA35)-L36-
T37-(AA38)-(AA39)-L40-T41-F42-K43-F44-Y45-M46-P47-K48-K49-
A50-T51-E52-L53-K54-(AA55)-L56-Q57-C58-L59-E60-E61-E62-L63-
20 K64-P65-L66-E67-E68-(AA69)-L70-N71-L72-A73-(AA74)-S75-K76-
N77-F78-H79-(AA80)-(AA81)-P82-R83-D84-(AA85)-(AA86)-S87-N88-
(AA89)-N90-V91-(AA92)-V93-L94-E95-L96-(AA97)-G98-S99-E100-
T101-T102-F103-(AA104)-C105-E106-Y107-A108-(AA109)-E110-T111-
A112-(AA113)-I114-V115-E116-F117-L118-N119-R120-W121-I122-
25 T123-F124-(AA125)-(AA126)-S127-I128-I129-(AA130)-T131-L132-T133

wherein:

- AA18 is R, L (wild type) G, M, F, E, W, K, Q, S, V, I, Y, H, D or T;
- AA22 is R, Q (wild type) E, G, A, L, M, W, K, S, V, I, Y, H, N, D, T, or F; and
- AA126 is H, Q (wild type) M, K, C, D, E, G, I, R, S, or T;

30 and optionally including one or more amino acid substitutions or deletions selected from the group consisting of:

- AA1 is A (wild type, a=1) or deleted (a=0);
 - AA2 is P (wild type, b=1) or deleted (b=0);
 - AA3 is T (wild type, c=1), C, A, G, Q, E, N, D, R, K, P, or deleted (c=0);
 - AA4 is S (wild type, d=1) or deleted (d=0);
 - 5 • AA5 is S (wild type, e=1) or deleted (e=0);
 - AA6 is S (wild type, f=1) or deleted (f=0);
 - AA7 is T (wild type, g=1) or deleted (g=0);
 - AA8 is K (wild type, h=1) or deleted (h=0);
 - AA9 is K (wild type, i=1) or deleted (i=0);
 - 10 • AA35 is K (wildtype) or E;
 - AA38 is R (wild type), W or G;
 - AA39 is M (wildtype), L or V;
 - AA55 is H (wildtype) or Y;
 - AA69 is V (wildtype) or A;
 - 15 • AA74 is Q (wild type), P, N, H, S;
 - AA80 is L (wild type), F or V;
 - AA81 is R (wild type), I, D or T;
 - AA85 is L (wild type) or V;
 - AA86 is I (wild type) or V;
 - 20 • AA89 is I (wild type) or V;
 - AA92 is I (wild type) or F;
 - AA97 is K (wild type) or Q;
 - AA104 is M (wild type) or A;
 - AA109 is D (wildtype), C or a non-natural amino acid with an activated side
 - 25 chain;
 - AA113 is T (wild type) or N;
 - AA125 is C (wild type), A or S; and
 - AA130 is S (wild type), T, G or R.; and
- with the proviso that if AA18 is R and AA22 is E, then AA126 is not H, M, K, C, D,
- 30 E, G, I, R, S, or T and the proviso that AA18, AA22 and AA126 are not all wild type.

In another aspect, the present disclosure provides a polypeptide comprising an amino acid sequence of the formula (SEQ ID NO: 97):

(AA1)_a-(AA2)_b-(AA3)_c-(AA4)_d-(AA5)_e-(AA6)_f-(AA7)_g-(AA8)_h-(AA9)_i-T10-Q11-L12-Q13-L14-E15-H16-L17-(AA18)-L19-D20-L21-(AA22)-M23-I24-L25-N26-G27-I28-N29-N30-
5 Y31-K32-N33-P34-(AA35)-L36-T37-(AA38)-(AA39)-L40-T41-F42-K43-F44-Y45-M46-P47-K48-K49-A50-T51-E52-L53-K54-(AA55)-L56-Q57-C58-L59-E60-E61-E62-L63-K64-P65-L66-E67-E68-(AA69)-L70-N71-L72-A73-(AA74)-S75-K76-N77-F78-H79-(AA80-(AA81)-P82-R83-D84-(AA85)-(AA86)-S87-N88-(AA89)-N90-(AA91)-(AA92)-V93-L94-E95-L96-(AA97)-G98-S99-E100-T101-T102-F103-(AA104)-C105-E106-Y107-A108-
10 (AA109)-E110-T111-A112-(AA113)-I114-V115-E116-F117-L118-N119-R120-W121-I122-T123-F124-(AA125)-(AA126)-S127-I128-I129-(AA130)-T131-L132-T133

wherein:

- each of a, b, c, d, e, f, g, h, and i is individually selected from 0 or 1;
- AA1 is A (wild type, a=1) or deleted (a=0);
- 15 • AA2 is P (wild type, b=1) or deleted (b=0);
- AA3 is T (wild type, c=1), C, A, G, Q, E, N, D, R, K, P, or deleted (c=0);
- AA4 is S (wild type, d=1) or deleted (d=0);
- AA5 is S (wild type, e=1) or deleted (e=0);

- AA6 is S (wild type, f=1) or deleted (f=0);
- AA7 is T (wild type, g=1) or deleted (g=0);
- AA8 is K (wild type, h=1) or deleted (h=0);
- AA9 is K (wild type, i=1) or deleted (i=0);
- AA18 is L (wild type) or R, L, G, M, F, E, H, W, K, Q, S, V, I, Y, H, D or T;
- AA22 is Q (wild type) or F, E, G, A, L, M, F, W, K, S, V, I, Y, H, R, N, D, T, or F;
- AA35 is K (wildtype) or E;
- AA38 is R (wild type), W or G;
- AA39 is M (wildtype), L or V;
- AA55 is H (wildtype) or Y;
- AA69 is V (wildtype) or A;
- AA74 is Q (wild type), P, N, H, S;
- AA80 is L (wild type), F or V;
- AA81 is R (wild type), I, D or T;
- AA85 is L (wild type) or V;
- AA86 is I (wild type) or V;
- AA89 is I (wild type) or V;
- AA91 is V (wild type), R or K;
- AA92 is I (wild type) or F;
- AA97 is K (wild type) or Q;
- AA104 is M (wild type) or A;
- AA109 is D (wildtype), C or a non-natural amino acid with an activated side chain;
- AA113 is T (wild type) or N;
- AA125 is C (wild type), A or S;
- AA126 is Q (wild type) or H, M, K, C, D, E, G, I, R, S, or T; and
- AA130 is S (wild type), T, G or R.; and

In another aspect, the disclosure provides a recombinant expression vector comprising the nucleic acid described herein. In some embodiments, the vector is a viral vector. In certain embodiments, the vector is a non-viral vector.

5 In another aspect, the disclosure provides a host cell transformed with a vector described herein.

In another aspect, the disclosure provides a pharmaceutical formulation comprising a polypeptide, a nucleic acid, or a vector described herein.

In another aspect, the disclosure provides the pharmaceutical formulation as described herein for use in therapy.

10 In another aspect, the disclosure provides a use of the pharmaceutical formulation as described herein in the manufacture of a medicament for treating a mammalian subject suffering from an autoimmune or inflammatory disease, disorder or condition or viral infection, wherein the pharmaceutical formulation as described herein is to be administered.

15 In another aspect, the disclosure provides a method of treating a mammalian subject suffering from an autoimmune or inflammatory disease, disorder or condition or viral infection, the method comprising administering a therapeutically-effective amount of pharmaceutical formulation described herein.

In some embodiments, the method further comprises administering one or more supplementary agents selected from the group consisting of a corticosteroid, a Janus kinase
20 inhibitor, a calcineurin inhibitor, a mTor inhibitor, an IMDH inhibitor, a biologic, a vaccine, and a therapeutic antibody. In certain embodiments, the therapeutic antibody is an antibody that binds a protein selected from the group consisting of BlyS, CD11a, CD20, CD25, CD3, CD52, IgEIL-12/IL-23, IL-17a, IL-1 β , IL-4R α , IL-5, IL-6R, integrin- α 4 β 7, RANKL, TNF α , VEGF-A, and VLA-4.

25 In some embodiments, the disease, disorder or condition is selected from viral infections, heliobacter pylori infection, HTLV, organ rejection, graft versus host disease, autoimmune thyroid disease, multiple sclerosis, allergy, asthma, neurodegenerative diseases including Alzheimer's disease, systemic lupus erythramatosis (SLE), autoinflammatory diseases, inflammatory bowel disease (IBD), Crohn's disease, diabetes, cartilage
30 inflammation, arthritis, rheumatoid arthritis, juvenile arthritis, juvenile rheumatoid arthritis, juvenile rheumatoid arthritis, polyarticular juvenile rheumatoid arthritis, systemic onset

juvenile rheumatoid arthritis, juvenile ankylosing spondylitis, juvenile enteropathic arthritis,
juvenile reactive arthritis, juvenile Reiter's Syndrome, SEA Syndrome, juvenile
dermatomyositis, juvenile psoriatic arthritis, juvenile scleroderma, juvenile systemic lupus
erythematosus, juvenile vasculitis, pauciarticular rheumatoidarthritis, polyarticular
5 rheumatoidarthritis, systemic onset rheumatoidarthritis, ankylosing spondylitis, enteropathic
arthritis, reactive arthritis, Reiter's syndrome, SEA Syndrome, psoriasis, psoriatic arthritis,

(AA1)_a-(AA2)_b-(AA3)_c-(AA4)_d-(AA5)_e-(AA6)_f-(AA7)_g-(AA8)_h-
 (AA9)_i-T10-Q11-L12-Q13-L14-E15-H16-L17-(AA18)-L19-D20-
 L21-(AA22)-M23-I24-L25-N26-G27-I28-N29-N30-Y31-K32-N33-
 P34-(AA35)-L36-T37-(AA38)-(AA39)-L40-T41-F42-K43-F44-
 Y45-M46-P47-K48-K49-A50-T51-E52-L53-K54-(AA55)-L56-
 Q57-C58-L59-E60-E61-E62-L63-K64-P65-L66-E67-E68-(AA69)-
 L70-N71-L72-A73-(AA74)-S75-K76-N77-F78-H79-(AA80-
 (AA81)-P82-R83-D84-(AA85)-(AA86)-S87-N88-(AA89)-N90-
 (AA91)-(AA92)-V93-L94-E95-L96-(AA97)-G98-S99-E100-T101-
 T102-F103-(AA104)-C105-E106-Y107-A108-(AA109)-E110-
 T111-A112-(AA113)-I114-V115-E116-F117-L118-N119-R120-
 W121-I122-T123-F124-(AA125)-(AA126)-S127-I128-I129-
 (AA130)-T131-L132-T133

Formula 1 (SEQ ID NO: 97)

wherein:

- each of a, b, c, d, e, f, g, h, and i is individually selected from 0 or 1;
- AA1 is A (wild type, a=1) or deleted (a=0);
- AA2 is P (wild type, b=1) or deleted (b=0);
- AA3 is T (wild type, c=1), C, A, G, Q, E, N, D, R, K, P, or deleted (c=0);
- AA4 is S (wild type, d=1) or deleted (d=0);
- AA5 is S (wild type, e=1) or deleted (e=0);
- AA6 is S (wild type, f=1) or deleted (f=0);
- AA7 is T (wild type, g=1) or deleted (g=0);
- AA8 is K (wild type, h=1) or deleted (h=0);
- AA9 is K (wild type, i=1) or deleted (i=0);
- AA18 is L (wild type) or R, L, G, M, F, E, H, W, K, Q, S, V, I, Y, H, D or T;
- AA22 is Q (wild type) or F, E, G, A, L, M, F, W, K, S, V, I, Y, H, R, N, D, T, or F;
- AA35 is K (wildtype) or E;
- AA38 is R (wild type), W or G;
- AA39 is M (wildtype), L or V;
- AA55 is H (wildtype) or Y;

- AA69 is V (wildtype) or A;
- AA74 is Q (wild type), P, N, H, S;
- AA80 is L (wild type), F or V;
- AA81 is R (wild type), I, D or T;
- AA85 is L (wild type) or V;
- AA86 is I (wild type) or V;
- AA89 is I (wild type) or V;
- AA91 is V (wild type), R or K;
- AA92 is I (wild type) or F;
- AA97 is K (wild type) or Q;
- AA104 is M (wild type) or A;
- AA109 is D (wildtype), C or a non-natural amino acid with an activated side chain;
- AA113 is T (wild type) or N;
- AA125 is C (wild type), A or S;
- AA126 is Q (wild type) or H, M, K, C, D, E, G, I, R, S, or T; and
- AA130 is S (wild type), T, G or R.; and

with the proviso that if AA18 is R and AA22 is E, then AA126 is not H, M, K, C, D, E, G, I, R, S, or T.

In certain embodiments, the disclosure provides IL2 muteins comprising the following mutations:

- AA18 is selected from the group consisting of L (wild type) or R, L, G, M, F, E, H, W, K, Q, S, V, I, Y, H, D or T;
- AA22 is selected from the group consisting of Q (wild type) or F, E, G, A, L, M, F, W, K, S, V, I, Y, H, R, N, D, T, or F; and
- AA126 is selected from the group consisting of Q (wild type) or H, M, K, C, D, E, G, I, R, S, or T,

with the proviso that if AA18 is R and AA22 is E, then AA126 is not H, M, K, C, D, E, G, I, R, S, or T.

In certain embodiments, the disclosure provides IL2 muteins comprising the following mutations:

- a=0;
- AA18 is selected from the group consisting of L (wild type) or R, L, G, M, F, E, H, W, K, Q, S, V, I, Y, H, D or T;

known in the art can be used for this purpose. The type of cross-linking reagent to use depends on the nature of the molecule to be coupled to IL-2 mutein and can readily be identified by those skilled in the art. Alternatively, or in addition, the IL2 mutein and/or the molecule to which it is intended to be conjugated may be chemically derivatized such that the two can be conjugated in a separate reaction as is also well known in the art.

Flag Tags

In other embodiments, the IL2 mutein can be modified to include an additional polypeptide sequence that functions as an antigenic tag, such as a FLAG sequence. FLAG sequences are recognized by biotinylated, highly specific, anti-FLAG antibodies, as described herein (see e.g., Blancar et al. (1992) Science 256:1014 and LeClair, et al. (1992) PNAS-USA 89:8145). In some embodiments, the IL2 mutein polypeptide further comprises a C-terminal c-myc epitope tag.

His Tags

In some embodiment, the IL2 muteins (including fusion proteins of such IL2 muteins) of the present invention are expressed as a fusion protein with one or more transition metal chelating polypeptide sequences. The incorporation of such a transition metal chelating domain facilitates purification immobilized metal affinity chromatography (IMAC) as described in Smith, et al. United States Patent No. 4,569,794 issued February 11, 1986. Examples of transition metal chelating polypeptides useful in the practice of the present invention are described in Smith, et al. *supra* and Dobeli, et al. United States Patent No. 5,320,663 issued May 10, 1995, the entire teachings of which are hereby incorporated by reference. Particular transition metal chelating polypeptides useful in the practice of the present invention are peptides comprising 3-6 contiguous histidine residues (SEQ ID NO: 98) such as a six-histidine peptide (His)₆ (SEQ ID NO: 99) and are frequently referred to in the art as “His-tags.”

Targeting Moieties:

In some embodiments, the IL2 mutein is provided as a fusion protein with a polypeptide sequence (“targeting domain”) to facilitate selective binding to particular cell type or tissue expressing a cell surface molecule that specifically binds to such targeting domain. In some embodiments, the targeting domain is an antibody (in particular a single domain antibody, scFv or VHH) or ligand that specifically binds to the surface protein of a

complementary nucleotides that overlap one another, but blunt-ended fragments can also be ligated. PCR-generated nucleic acids can also be used to generate various mutant sequences.

An IL2 mutein of the present disclosure may be produced recombinantly not only directly, but also as a fusion polypeptide with a heterologous polypeptide, *e.g.* a signal sequence or other polypeptide having a specific cleavage site at the N-terminus or C-terminus of the mature IL2 mutein. In general, the signal sequence may be a component of the vector, or it may be a part of the coding sequence that is inserted into the vector. The heterologous signal sequence selected preferably is one that is recognized and processed (*i.e.*, cleaved by a signal peptidase) by the host cell. In some embodiments, the signal sequence is the signal sequence that is natively associated with the IL2 mutein (*i.e.* the human IL2 signal sequence). The inclusion of a signal sequence depends on whether it is desired to secrete the IL-2 mutein from the recombinant cells in which it is made. If the chosen cells are prokaryotic, it generally is preferred that the DNA sequence not encode a signal sequence. If the chosen cells are eukaryotic, it generally is preferred that a signal sequence be encoded and most preferably that the wild type IL-2 signal sequence be used. Alternatively, heterologous mammalian signal sequences may be suitable, such as signal sequences from secreted polypeptides of the same or related species, as well as viral secretory leaders, for example, the herpes simplex gD signal. When the recombinant host cell is a yeast cell such as *Saccharomyces cerevisiae*, the alpha mating factor secretion signal sequence may be employed to achieve extracellular secretion of the IL2 mutein into the culture medium as described in Singh, United States Patent No. 7,198,919 B1 issued April 3, 2007.

In the event the IL2 mutein to be expressed is to be expressed as a chimera (*e.g.*, a fusion protein comprising an IL2 mutein and a heterologous polypeptide sequence), the chimeric protein can be encoded by a hybrid nucleic acid molecule comprising a first sequence that encodes all or part of the IL-2 mutein and a second sequence that encodes all or part of the heterologous polypeptide. For example, subject IL-2 muteins described herein may be fused to a hexa-histidine tag (SEQ ID NO: 99) to facilitate purification of bacterially expressed protein, or to a hemagglutinin tag to facilitate purification of protein expressed in eukaryotic cells. By first and second, it should not be understood as limiting to the orientation of the elements of the fusion protein and a heterologous polypeptide can be linked at either the N-terminus and/or C-terminus of the IL2 mutein. For example, the N-terminus may be linked to a targeting domain and the C-terminus linked to a hexa-histidine tag (SEQ ID NO: 99) purification handle.

EXAMPLES

The following examples are provided to describe certain embodiments of the invention provided herein and are not to be construed to as limiting.

Example 1: Generation of the human IL2 expression vector pcDNA3.1/hygro(+)-huIL2

The human IL2 DNA open reading frame (“ORF”) (Genbank NM_000586.3) was synthesized (Life Technologies GeneArt Service, Carlsbad, CA), and amplified via PCR using Platinum SuperFi II DNA polymerase kit (commercially available as catalog #12361050, ThermoFisher) in substantial accordance with the manufacturer’s protocol, and using primers 5’ TATAGTCAGCGCCACcCATGTACAGGATGCAACTCCTGTC 3’ (SEQ ID NO: 100), which incorporates an NheI restriction site, and 5’ TATAGGGCCCTATCAAGTCAGTGTTGAGATG 3’ (SEQ ID NO: 101), which incorporates an ApaI restriction site. The PCR fragment was visualized on a 1% agarose gel (item #54803, Lonza, Rockland, ME), excised from the gel and purified using a QIAquick PCR Purification kit (commercially available as catalog #28106, Qiagen, Germany) according to the manufacturer’s protocol.

The purified PCR fragment and mammalian expression vector pcDNA 3.1/Hygro(+) (commercially available as catalog #V87020, ThermoFisher, Carlsbad CA) were digested with NheI and ApaI (commercially available as catalog #R0111S and #R0114L, New England Biolabs, Ipswich, MA) restriction enzymes. The expression vector was further treated with a Quick Dephosphorylation kit (commercially available as catalog #M0508L, New England Biolabs) in substantial accordance with the manufacturer’s protocol. The PCR fragment was ligated into pcDNA 3.1/Hygro(+) using the Rapid DNA Ligation Kit (commercially available as catalog #11635379001, Sigma Aldrich, St. Louis, MO) in substantial accordance with the manufacturer’s protocol, transformed into One Shot TOP10 Chemically Competent E. coli (commercially available as catalog #C404006, Life Technologies, Carlsbad, CA), plated onto LB Agar plates containing 100ug/ml carbenicillin (commercially available as catalog #L1010, Teknova, Hollister, CA), and grown overnight at 37C.

The following day individual bacterial colonies were picked and used to start a 3ml bacterial culture in LB Broth (#10855-001, Life Technologies) with 100ug/ml ampicillin (commercially available as catalog #A9626, Teknova). The cultures were grown overnight at

CLAIMS

1. A polypeptide having decreased binding affinity, compared to a wild type hIL2, to the extracellular domain of hCD132, the polypeptide comprising an amino acid sequence of the formula:

5 (AA1)_a-(AA2)_b-(AA3)_c-(AA4)_d-(AA5)_e-(AA6)_f-(AA7)_g-(AA8)_h-(AA9)_i-
T10-Q11-L12-Q13-L14-E15-H16-L17-(AA18)-L19-D20-L21-(AA22)-
M23-I24-L25-N26-G27-I28-N29-N30-Y31-K32-N33-P34-(AA35)-L36-
T37-(AA38)-(AA39)-L40-T41-F42-K43-F44-Y45-M46-P47-K48-K49-
A50-T51-E52-L53-K54-(AA55)-L56-Q57-C58-L59-E60-E61-E62-L63-
10 K64-P65-L66-E67-E68-(AA69)-L70-N71-L72-A73-(AA74)-S75-K76-
N77-F78-H79-(AA80)-(AA81)-P82-R83-D84-(AA85)-(AA86)-S87-N88-
(AA89)-N90-V91-(AA92)-V93-L94-E95-L96-(AA97)-G98-S99-E100-
T101-T102-F103-(AA104)-C105-E106-Y107-A108-(AA109)-E110-T111-
A112-(AA113)-I114-V115-E116-F117-L118-N119-R120-W121-I122-
15 T123-F124-(AA125)-(AA126)-S127-I128-I129-(AA130)-T131-L132-T133

wherein:

- AA18 is R, L (wild type) G, M, F, E, W, K, Q, S, V, I, Y, H, D or T;
- AA22 is R, Q (wild type) E, G, A, L, M, W, K, S, V, I, Y, H, N, D, T, or F; and
- AA126 is H, Q (wild type) M, K, C, D, E, G, I, R, S, or T;

20 and optionally including one or more amino acid substitutions or deletions selected from the group consisting of:

- AA1 is A (wild type, a=1) or deleted (a=0);
- AA2 is P (wild type, b=1) or deleted (b=0);
- AA3 is T (wild type, c=1), C, A, G, Q, E, N, D, R, K, P, or deleted (c=0);
- 25 • AA4 is S (wild type, d=1) or deleted (d=0);
- AA5 is S (wild type, e=1) or deleted (e=0);
- AA6 is S (wild type, f=1) or deleted (f=0);
- AA7 is T (wild type, g=1) or deleted (g=0);
- AA8 is K (wild type, h=1) or deleted (h=0);
- 30 • AA9 is K (wild type, i=1) or deleted (i=0);
- AA35 is K (wildtype) or E;

- AA38 is R (wild type), W or G;
- AA39 is M (wildtype), L or V;
- AA55 is H (wildtype) or Y;
- AA69 is V (wildtype) or A;
- 5 • AA74 is Q (wild type), P, N, H, S;
- AA80 is L (wild type), F or V;
- AA81 is R (wild type), I, D or T;
- AA85 is L (wild type) or V;
- AA86 is I (wild type) or V;
- 10 • AA89 is I (wild type) or V;
- AA92 is I (wild type) or F;
- AA97 is K (wild type) or Q;
- AA104 is M (wild type) or A;
- AA109 is D (wildtype), C or a non-natural amino acid with an activated side
- 15 chain;
- AA113 is T (wild type) or N;
- AA125 is C (wild type), A or S; and
- AA130 is S (wild type), T, G or R.; and

with the proviso that if AA18 is R and AA22 is E, then AA126 is not H, M, K, C, D, E, G, I,
 20 R, S, or T and the proviso that AA18, AA22 and AA126 are not all wild type.

2. The polypeptide of claim 1 wherein the polypeptide comprises one or more
 amino acid substitutions selected from positions 18, 22 and 126 numbered in accordance with
 mature wild type hIL-2, wherein

- 25 • the amino acid substitution at position 18 is selected from the group consisting
 of R, G, M, F, E, W, K, Q, S, V, I, Y, H, D and T;
- the amino acid substitution at position 22 is selected from the group consisting
 of R, E, G, A, L, M, W, K, S, V, I, Y, H, N, D, T, and F; and
- the amino acid substitution at position 126 is selected from the group
- 30 consisting of H, M, K, C, D, E, G, I, R, S, and T,
 optionally wherein the polypeptide comprises the amino acid substitutions at
 amino acid positions 18, 22 and 126.

3. The polypeptide of claim 1 or 2 wherein a=0, optionally wherein each of a-i = 0.

5 4. The polypeptide of any one of claims 1-3 wherein said polypeptide comprises a set of mutations selected from the group consisting of: L18R, Q22R and Q126H; L18R and Q126H; Q22E and Q126H; L18G, Q22E and Q126H; L18A, Q22E and Q126H; L18M, Q22E and Q126H; L18F, Q22E and Q126H; L18W, Q22E and Q126H; L18K, Q22E and Q126H; L18Q, Q22E and Q126H; L18E, Q22E and Q126H; L18S, Q22E and Q126H; L18V, Q22E and Q126H; L18I, Q22E and Q126H; L18Y, Q22E and Q126H; L18H, Q22E and Q126H; L18N, Q22E and Q126H; L18D, Q22E and Q126H; L18T, Q22E and Q126H; L18R, Q22G and Q126H; L18R, Q22A and Q126H; L18R, Q22L and Q126H; L18R, Q22M and Q126H; L18R, Q22F and Q126H; L18R, Q22W and Q126H; L18R, Q22K and Q126H; L18R, Q22S and Q126H; L18R, Q22V and Q126H; L18R, Q22I and Q126H; L18R Q22Y and Q126H; L18R Q22H and Q126H; L18R, Q22N and Q126H; L18R, Q22D and Q126H; and L18R, Q22T and Q126H.

5. The polypeptide of any one of claims 1-4, wherein the polypeptide comprises the sequence:

20 TQLQLEHLRLDLRMILNGINNYKNPKLTRMLTFKFYMPKKATELKHLQCLEEELKPLEEVLNLAQSKNFHLRPRDLISNINVIVLELKGSETTFMCEYADETATIVEFLNRWITFC HSIISTLT;

or

25 APTSSSTKKTQLQLEHLRLDLRMILNGINNYKNPKLTRMLTFKFYMPKKATELKHLQCLEEELKPLEEVLNLAQSKNFHLRPRDLISNINVIVLELKGSETTFMCEYADETATIVE FLNRWITFC HSIISTLT.

6. The polypeptide of any one of claims 1-5 wherein the polypeptide is PEGylated.

7. The polypeptide of any one of claims 1-5 wherein the polypeptide is PEGylated and the PEG component of such PEGylated polypeptide has a molecular weight of from about 10kD to about 70kD.

8. The polypeptide of any one of claims 1-7 wherein the polypeptide is a fusion protein.

9. The polypeptide of Claim 8 wherein the fusion protein comprises an Fc domain.
10. A nucleic acid encoding a polypeptide of any one of claims 1-9.
11. The nucleic acid of claim 10 wherein the nucleic acid is DNA.
- 5 12. A recombinant expression vector comprising the nucleic acid of claim 10 or 11.
13. The vector of claim 12 wherein said vector is a viral vector.
14. The vector of claim 12 wherein said vector is a non-viral vector.
15. A host cell transformed with a vector of any one of claims 12-14.
- 10 16. A pharmaceutical formulation comprising a polypeptide of any one of claims 1-8, the nucleic acids of claims 9 and 10 or a vector of claims 11-13.
17. Use of the pharmaceutical formulation of claim 16 in the manufacture of a medicament for treating a mammalian subject suffering from an autoimmune or inflammatory disease, disorder or condition or viral infection, wherein the pharmaceutical
- 15 formulation is to be administered.
18. The use of claim 17, wherein one or more supplementary agents selected from the group consisting of a corticosteroid, a Janus kinase inhibitor, a calcineurin inhibitor, a mTor inhibitor, an IMDH inhibitor, a biologic, a vaccine, and a therapeutic antibody is to be further administered.
- 20 19. The use of claim 18, wherein the therapeutic antibody is an antibody that binds a protein selected from the group consisting of BLYS, CD11a, CD20, CD25, CD3, CD52, IgEIL-12/IL-23, IL-17a, IL-1 β , IL-4R α , IL-5, IL-6R, integrin- α 4 β 7, RANKL, TNF α , VEGF-A, and VLA-4.
- 25 20. The use of any one of claims 17-19, wherein the disease, disorder or condition is selected from viral infections, heliobacter pylori infection, HTLV, organ rejection, graft versus host disease, autoimmune thyroid disease, multiple sclerosis, allergy, asthma, neurodegenerative diseases including Alzheimer's disease, systemic lupus erythramatosis (SLE), autoinflammatory diseases, inflammatory bowel disease (IBD), Crohn's disease, diabetes, cartilage inflammation, arthritis, rheumatoid arthritis, juvenile arthritis, juvenile
- 30 rheumatoid arthritis, juvenile rheumatoid arthritis, polyarticular juvenile rheumatoid arthritis, systemic onset juvenile rheumatoid arthritis, juvenile ankylosing spondylitis, juvenile enteropathic arthritis, juvenile reactive arthritis, juvenile Reiter's Syndrome, SEA Syndrome,

juvenile dermatomyositis, juvenile psoriatic arthritis, juvenile scleroderma, juvenile systemic lupus erythematosus, juvenile vasculitis, pauciarticular rheumatoidarthritis, polyarticular rheumatoidarthritis, systemic onset rheumatoidarthritis, ankylosing spondylitis, enteropathic arthritis, reactive arthritis, Reiter's syndrome, SEA Syndrome, psoriasis, psoriatic arthritis, 5 dermatitis (eczema), exfoliative dermatitis or atopic dermatitis, pityriasis rubra pilaris, pityriasis rosacea, parapsoriasis, pityriasis lichenoides, lichen planus, lichen nitidus, ichthyosiform dermatosis, keratodermas, dermatosis, alopecia areata, pyoderma gangrenosum, vitiligo, pemphigoid, urticaria, prokeratosis, rheumatoid arthritis; seborrheic dermatitis, solar dermatitis; seborrheic keratosis, senile keratosis, actinic keratosis, photo- 10 induced keratosis, keratosis follicularis; acne vulgaris; keloids; nevi; warts including verruca, condyloma or condyloma acuminatum, and human papilloma viral (HPV) infections.

21. The pharmaceutical formulation of claim 16 for use in therapy.