

ANTI-SIRP-ALPHA ANTIBODIES

SEQUENCE LISTING

[0001] The instant application contains a Sequence Listing which has been submitted electronically in ASCII format and is hereby incorporated by reference in its entirety. Said ASCII copy, created on May 6, 2022, is named 105218-03-5011-WO_Sequence_Listing.txt and is 453,000 bytes in size.

RELATED APPLICATION DISCLOSURE

[0002] The subject application claims the benefit of U.S. Provisional Application No. 63/197,259, filed June 4, 2021, U.S. Provisional Application No. 63/325,828, filed March 31, 2022, and 63/339,326, filed May 6, 2022, each of which is hereby incorporated by reference in its entirety.

FIELD OF THE INVENTION

[0003] This invention generally relates to anti-SIRP α (Signal regulatory protein alpha) antibodies or antigen-binding fragments thereof for therapeutic and/or diagnostic use. More specifically, the invention relates to anti-SIRP α antibodies or antigen-binding fragments thereof and methods of their use for the treatment of various diseases or disorders, for example, cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease, or fibrosis. Pharmaceutical compositions and kits comprising the anti-SIRP α antibodies or antigen-binding fragments thereof are also disclosed.

BACKGROUND OF THE INVENTION

[0004] SIRP α is an inhibitory receptor expressed on myeloid cells such as macrophages, neutrophils and subsets of dendritic cells. SIRP α contains three Ig-like domains, a single transmembrane domain, and a cytoplasmic tail with four tyrosine residues which form two typical immunoreceptor tyrosine based inhibitory motifs (ITIMs). The natural ligand for SIRP α is CD47, expressed on many cells including erythrocytes and platelets. Binding of SIRP α to CD47 leads to the phosphorylation of the tyrosine residues in SIRP α intracellular ITIM domain and subsequent recruitment and activation SHP-1 and SHP-2 phosphatases at the cell membrane which can then,

by dephosphorylation of downstream targets, regulate cellular functions including phagocytosis or antigen presentation.

[0005] The development of an effective SIRP α antagonist is complicated by polymorphisms within the CD47 binding domain. It has been reported that there may be up to ten allelic variants in the general population (Takenaka 2007; *Nat Immunol* 2007 Dec;8(12):1313-23. doi: 10.1038/ni1527.) and recent studies (Treffers, 2018, *Eur J Immunol*. 2018 Feb;48(2):344-354. doi: 10.1002/eji.201747215.; *MAbs* Aug/Sep 2019;11(6):1036-1052. doi: 10.1080/19420862.2019.1624123.) highlight that two SIRP α variants, V1 and V2, constitute the most prevalent allelic groups: homozygous V1/V1, homozygous V2/V2, and heterozygous V1/V2. These variants differ in 13 out of 118 amino acid residues in the N-terminal immunoglobulin-like domain of SIRP α responsible for CD47 binding. These polymorphic residues are located outside the CD47 binding site and, accordingly, the affinity of CD47 binding to SIRP α variants is similar (Hatherley D, 2008 *Immunity* 2008 Nov 14;29(5):675-8. doi: 10.1016/j.immuni.2008.10.004.). Consequently, therapeutic targeting of SIRP α in diverse patient population irrespective of SIRP α genotype necessitates pan-allelic antibodies that cross-react with the two major SIRP α alleles (V1 and V2).

[0006] In addition to considering polymorphic variants when targeting SIRP α , one also should consider SIRP α 's closest relatives, SIRP β 1 and SIRP γ given their high sequence conservation particularly in N-terminal domains. SIRP β 1, like SIRP α is also expressed predominantly on cells of the myeloid lineage, but unlike SIRP α , lacks its own signaling cytoplasmic domain, but harbors a positively charged amino acid residue within the transmembrane region allowing for the stable association with ITAM-containing adapter molecule DAP12 and therefore is presumed to act as an activating receptor. SIRP β 1 does not bind CD47, and its ligands have not been identified. There are at least two isoforms of SIRP β 1 (Liu et al 2007 *J Mol Biol*. 2007 Jan 19;365(3):680-93. doi: 10.1016/j.jmb.2006.09.079; Brooke et al. 2004 *J Immunol*. 2004 Aug 15;173(4):2562-70. doi: 10.4049/jimmunol.173.4.2562.) that arose through tandem duplication of the gene within the SIRP family gene cluster. SIRP γ is exclusively expressed on T-cells and activated NK cells and does bind to CD47 with 10-fold lower affinity than SIRP α :CD47 interaction. Although it does not have intrinsic signaling capacity, there is reported evidence that it plays a role in T-cell transendothelial migration (TEM) and antigen presentation.

[0007] In view of the above, there is a need for pan-allelic anti-SIRP α antibodies capable of blocking the interaction of SIRP α with CD47 and that are selective against SIRP γ to allow for

treatment of several types of cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease, or fibrosis.

BRIEF SUMMARY OF THE INVENTION

[0008] The present invention addresses the above need by providing agents, in particular antibodies or antigen-binding fragments thereof, which bind (e.g., specifically) to SIRP α , in particular human SIRP α . In one aspect of the invention, the antibodies or antigen-binding fragments thereof, block the interaction between SIRP α and CD47. In another aspect of the invention, the antibodies or antigen-binding fragments thereof, block CD47-mediated SIRP α signaling.

[0009] The antibodies or antigen-binding fragments thereof of the invention are useful, for example, for the treatment and/or prevention of diseases or disorders that can be alleviated by modulating the interaction between SIRP α and CD47, in particular by blocking CD47-mediated SIRP α signaling. In one aspect of the invention, the antibodies or antigen-binding fragments thereof, are useful, for example for the treatment of cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis, preferably cancer.

[0010] The invention provides an anti-SIRP α antibody or antigen-binding fragments thereof, having one or more of the properties described herein below.

[0011] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof binds specifically to SIRP α , in particular human SIRP α or cynomolgus monkey SIRP α , more particularly human SIRP α . In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof binds to the V1 and/or V2 alleles of human SIRP α . In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof does not bind (e.g., there is no detectable binding or the anti-SIRP α antibody or antigen-binding fragment thereof binds with a K_D of 1 μ M or greater) to SIRP γ , in particular cynomolgus monkey SIRP γ or human SIRP γ , more particularly human SIRP γ . In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, does not bind (e.g., there is no detectable binding or the anti-SIRP α antibody or antigen-binding fragment thereof binds with a K_D of 1 μ M or greater) to rabbit, mouse, rat, or dog SIRP α .

[0012] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof blocks the binding of CD47 to SIRP α , in particular human SIRP α or cynomolgus monkey

SIRP α , more particularly human SIRP α . In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof blocks the binding of CD47 to human SIRP α -V1 and SIRP α -V2. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof blocks CD47-mediated SIRP α signaling. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof enhances phagocytosis by macrophages and/or dendritic cells. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof enhances antibody dependent cellular phagocytosis (ADCP), in combination with a tumor targeting agent, in particular a tumor targeting antibody. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof enhances phagocytosis of tumor cells by macrophages and/or dendritic cells. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof blocks inhibition of T-cell proliferation. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof has favorable pharmacokinetic properties. In one aspect of the invention, an anti-SIRP α antibody or antigen-binding fragment thereof has favorable biophysical properties, for example yield, quality, stability or solubility.

[0013] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 34 (H-CDR2); and the amino acid sequence of SEQ ID NO: 35 (H-CDR3), and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 or SEQ ID NO: 37 (L-CDR1); the amino acid sequence of SEQ ID NO: 38 (L-CDR2); and the amino acid sequence of SEQ ID NO: 39 (L-CDR3),
or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 1 or SEQ ID NO: 223 (H-CDR1); the amino acid sequence of SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5 or SEQ ID NO: 224 (H-CDR2); and the amino acid sequence of SEQ ID NO: 6 (H-CDR3); and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9 or SEQ ID NO: 225 (L-CDR1); the amino acid sequence of SEQ ID NO: 10, SEQ ID NO: 11 or SEQ ID NO: 226 (L-CDR2); and the amino acid sequence of SEQ ID NO: 12 or SEQ ID NO: 227 (L-CDR3),
or

- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 52 (H-CDR1); the amino acid sequence of SEQ ID NO: 53 (H-CDR2); and the amino acid sequence of SEQ ID NO: 54 (H-CDR3); and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 55 (L-CDR1); the amino acid sequence of SEQ ID NO: 56 (L-CDR2); and the amino acid sequence of SEQ ID NO: 57 (L-CDR3),
or
- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 70 (H-CDR2); and the amino acid sequence of SEQ ID NO: 71 (H-CDR3); and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 (L-CDR1); the amino acid sequence of SEQ ID NO: 72 (L-CDR2); and the amino acid sequence of SEQ ID NO: 39 (L-CDR3),
or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 262 (H-CDR1); the amino acid sequence of SEQ ID NO: 87 (H-CDR2); and the amino acid sequence of SEQ ID NO: 88 (H-CDR3); and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 (L-CDR1); the amino acid sequence of SEQ ID NO: 72 (L-CDR2); and the amino acid sequence of SEQ ID NO: 89 (L-CDR3).

[0014] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 34 (H-CDR2); the amino acid sequence of SEQ ID NO: 35 (H-CDR3); and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 233, wherein amino acids X1 = D or G and X2 = L or A (L-CDR1); the amino acid sequence of SEQ ID NO: 38, (L-CDR2); the amino acid sequence of SEQ ID NO: 39 (L-CDR3), or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 228 (H-CDR1), wherein amino acids X1 = N or D; the amino acid sequence of SEQ ID NO: 229, wherein X1 = Y or D, X2 = N or T, X3 = N or Q, and X4 = S or P (H-CDR2); the amino acid sequence of SEQ ID NO: 6 (H-CDR3); and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 230, wherein X1 = K or R, X2 = N or T, X3 = G or A and X4 = N, A

or T (L-CDR1); the amino acid sequence of SEQ ID NO:231, wherein X1=L, Q or G and X2=N or S (L-CDR2); the amino acid sequence of SEQ ID NO:232, wherein X1=M or G (L-CDR3).

[0015] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 100, 110, 111, 112, 113, 114, 115, 116, or 117; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 105, 125, or 126.

[0016] In a further aspect, an anti-SIRP α antibody or an antigen-binding fragment thereof of the present invention comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 104, 118, 119, 120, 121, 122, 123, 124 or 221; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 109, 127, 128, 129, 130 or 222.

[0017] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 100, 101, 102, 103, or 104; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 105, 106, 107, 108, or 109.

[0018] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 100; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 105; or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 110; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 111; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 112; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 113; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- f) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 114; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- g) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 115; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- h) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 116; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or

- i) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 117; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- j) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 111; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126; or
- k) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 112; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126; or
- l) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 113; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126; or
- m) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 114; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126; or
- n) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 115; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126; or
- o) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 116; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126; or
- p) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 117; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126.

[0019] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 104; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 109; or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 118; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or
- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 118; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 128; or
- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 119; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 119; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129; or
- f) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 120; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or
- g) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 120; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129; or
- h) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 121; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or

- i) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 122; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or
- j) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 118; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- k) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 121; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129; or
- l) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 122; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129; or
- m) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 119; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- n) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 123; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or
- o) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 120; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- p) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 123; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129; or
- q) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 121; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- r) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 122; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- s) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 124; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129; or
- t) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 124; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127; or
- u) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 123; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- v) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 124; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130; or
- w) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 221; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 222.

[0020] In one aspect of the invention, an anti-SIRP α antibody or an antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 100; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 105; or

- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 101; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 106; or
- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 102; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 107; or
- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 103; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 108; or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 104; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 109.

[0021] In one aspect of the invention, an anti-SIRP α antibody comprises a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 148, 149, 150, 151 or 152; and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195 or 218.

[0022] In a further aspect, an anti-SIRP α antibody of the invention comprises:

- a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 131; and a light chain comprising the amino acid sequence of SEQ ID NO: 174; or
- b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 138; and a light chain comprising the amino acid sequence of SEQ ID NO: 121; or
- c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 139; and a light chain comprising the amino acid sequence of SEQ ID NO: 182; or
- d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 140; and a light chain comprising the amino acid sequence of SEQ ID NO: 183; or
- e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 141; and a light chain comprising the amino acid sequence of SEQ ID NO: 184; or
- f) a heavy chain comprising the amino acid sequence of SEQ ID NO: 142; and a light chain comprising the amino acid sequence of SEQ ID NO: 185; or
- g) a heavy chain comprising the amino acid sequence of SEQ ID NO: 143; and a light chain comprising the amino acid sequence of SEQ ID NO: 186; or
- h) a heavy chain comprising the amino acid sequence of SEQ ID NO: 144; and a light chain comprising the amino acid sequence of SEQ ID NO: 187; or
- i) a heavy chain comprising the amino acid sequence of SEQ ID NO: 145; and a light chain comprising the amino acid sequence of SEQ ID NO: 188; or

- j) a heavy chain comprising the amino acid sequence of SEQ ID NO: 146; and a light chain comprising the amino acid sequence of SEQ ID NO: 189; or
- k) a heavy chain comprising the amino acid sequence of SEQ ID NO: 147; and a light chain comprising the amino acid sequence of SEQ ID NO: 190; or
- l) a heavy chain comprising the amino acid sequence of SEQ ID NO: 148; and a light chain comprising the amino acid sequence of SEQ ID NO: 191; or
- m) a heavy chain comprising the amino acid sequence of SEQ ID NO: 149; and a light chain comprising the amino acid sequence of SEQ ID NO: 192; or
- n) a heavy chain comprising the amino acid sequence of SEQ ID NO: 150; and a light chain comprising the amino acid sequence of SEQ ID NO: 193; or
- o) a heavy chain comprising the amino acid sequence of SEQ ID NO: 151; and a light chain comprising the amino acid sequence of SEQ ID NO: 194; or
- p) a heavy chain comprising the amino acid sequence of SEQ ID NO: 152; and a light chain comprising the amino acid sequence of SEQ ID NO: 195; or
- q) a heavy chain comprising the amino acid sequence of SEQ ID NO: 217; and a light chain comprising the amino acid sequence of SEQ ID NO: 218.

[0023] In one aspect of the invention, an anti-SIRP α antibody comprises a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 135, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173 or 219; and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216 or 220.

[0024] In one aspect of the invention, an anti-SIRP α antibody comprises:

- a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 135; and a light chain comprising the amino acid sequence of SEQ ID NO: 178; or
- b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 153; and a light chain comprising the amino acid sequence of SEQ ID NO: 196; or
- c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 154; and a light chain comprising the amino acid sequence of SEQ ID NO: 197; or
- d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 155; and a light chain comprising the amino acid sequence of SEQ ID NO: 198; or
- e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 156; and a light chain comprising the amino acid sequence of SEQ ID NO: 199; or

- f) a heavy chain comprising the amino acid sequence of SEQ ID NO: 157; and a light chain comprising the amino acid sequence of SEQ ID NO: 200; or
- g) a heavy chain comprising the amino acid sequence of SEQ ID NO: 158; and a light chain comprising the amino acid sequence of SEQ ID NO: 201; or
- h) a heavy chain comprising the amino acid sequence of SEQ ID NO: 159; and a light chain comprising the amino acid sequence of SEQ ID NO: 202; or
- i) a heavy chain comprising the amino acid sequence of SEQ ID NO: 160; and a light chain comprising the amino acid sequence of SEQ ID NO: 203; or
- j) a heavy chain comprising the amino acid sequence of SEQ ID NO: 161; and a light chain comprising the amino acid sequence of SEQ ID NO: 204; or
- k) a heavy chain comprising the amino acid sequence of SEQ ID NO: 162; and a light chain comprising the amino acid sequence of SEQ ID NO: 205; or
- l) a heavy chain comprising the amino acid sequence of SEQ ID NO: 163; and a light chain comprising the amino acid sequence of SEQ ID NO: 206; or
- m) a heavy chain comprising the amino acid sequence of SEQ ID NO: 164; and a light chain comprising the amino acid sequence of SEQ ID NO: 207; or
- n) a heavy chain comprising the amino acid sequence of SEQ ID NO: 165; and a light chain comprising the amino acid sequence of SEQ ID NO: 208; or
- o) a heavy chain comprising the amino acid sequence of SEQ ID NO: 166; and a light chain comprising the amino acid sequence of SEQ ID NO: 209; or
- p) a heavy chain comprising the amino acid sequence of SEQ ID NO: 167; and a light chain comprising the amino acid sequence of SEQ ID NO: 210; or
- q) a heavy chain comprising the amino acid sequence of SEQ ID NO: 168; and a light chain comprising the amino acid sequence of SEQ ID NO: 211; or
- r) a heavy chain comprising the amino acid sequence of SEQ ID NO: 169; and a light chain comprising the amino acid sequence of SEQ ID NO: 212; or
- s) a heavy chain comprising the amino acid sequence of SEQ ID NO: 170; and a light chain comprising the amino acid sequence of SEQ ID NO: 213; or
- t) a heavy chain comprising the amino acid sequence of SEQ ID NO: 171; and a light chain comprising the amino acid sequence of SEQ ID NO: 214; or
- u) a heavy chain comprising the amino acid sequence of SEQ ID NO: 172; and a light chain comprising the amino acid sequence of SEQ ID NO: 215; or
- v) a heavy chain comprising the amino acid sequence of SEQ ID NO: 173; and a light chain comprising the amino acid sequence of SEQ ID NO: 216; or

w) a heavy chain comprising the amino acid sequence of SEQ ID NO: 219; and a light chain comprising the amino acid sequence of SEQ ID NO: 220.

[0025] In one aspect of the invention, an anti-SIRP α antibody comprises a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 131, 133, 134, 137 or 135; and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 174, 176, 177, 180, or 178.

[0026] In one aspect of the invention, an anti-SIRP α antibody comprises:

- a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 131; and a light chain comprising the amino acid sequence of SEQ ID NO: 174; or
- b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 133; and a light chain comprising the amino acid sequence of SEQ ID NO: 176; or
- c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 134; and a light chain comprising the amino acid sequence of SEQ ID NO: 177; or
- d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 137; and a light chain comprising the amino acid sequence of SEQ ID NO: 180; or
- e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 135; and a light chain comprising the amino acid sequence of SEQ ID NO: 178.

[0027] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, is a human antibody or antigen-binding fragment thereof. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, is a monoclonal antibody. In one aspect of the invention, the anti-SIRP α antibody is a full-length antibody. In one aspect of the invention, the anti-SIRP α antibody or fragment thereof, is a human monoclonal antibody, for example a full-length human monoclonal antibody.

[0028] In a one aspect of the invention, the anti-SIRP α antibody, or an antigen-binding fragment thereof, comprises a heavy chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 100, 110, 111, 112, 113, 114, 115, 116, or 117 and a light chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 105, 125, or 126. In a further aspect of the invention, the antibody or antigen-binding fragment thereof that comprises a heavy chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 100, 110, 111, 112, 113, 114, 115, 116, or 117 and a light chain variable region having at least 90%, at least 95%, at least 98%, or

at least 99% identity to the amino acid sequences of SEQ ID NO: 105, 125, or 126 specifically binds to SIRP α . In a one aspect of the invention, the anti-SIRP α antibody, or an antigen-binding fragment thereof, comprises a heavy chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 104, 118, 119, 120, 121, 122, 123, 124, or 221 and a light chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 109, 127, 128, 129, 130, or 222. In a further aspect of the invention, the antibody or antigen-binding fragment thereof that comprises a heavy chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 104, 118, 119, 120, 121, 122, 123, 124, or 221 and a light chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 109, 127, 128, 129, 130, or 222 specifically binds to SIRP α . In one aspect of the invention, the anti-SIRP α antibody, or an antigen-binding fragment thereof, comprises a heavy chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 101, 102 or 103 and a light chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 106, 107, 108. In a further aspect of the invention, the antibody or antigen-binding fragment thereof that comprises a heavy chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 101, 102 or 103 and a light chain variable region having at least 90%, at least 95%, at least 98%, or at least 99% identity to the amino acid sequences of SEQ ID NO: 106, 107, 108 specifically binds to SIRP α .

[0029] In one aspect of the invention, the anti-SIRP α antibody, or an antigen-binding fragment thereof, comprises a heavy chain constant region from an antibody selected from the group consisting of IgG1, IgG2, IgG3, IgG4, IgM, IgA and IgE, for example human IgG1, IgG2, IgG3, IgG4, IgM, IgA or IgE, more particularly, human IgG1, IgG2, IgG3, IgG4.

[0030] In one aspect of the invention, the anti-SIRP α antibody or an antigen-binding fragment thereof comprises a heavy chain constant region, wherein the heavy chain constant region is of an IgG4 with a S241P substitution. In one aspect of the invention, the anti-SIRP α antibody or an antigen-binding fragment thereof comprises a heavy chain lacking a C-terminal lysine residue.

[0031] In one aspect of the invention, the anti-SIRP α antibody or an antigen-binding fragment thereof comprises a heavy chain constant region wherein the heavy chain constant region is of an IgG1 with L234A and L235A substitutions.

[0032] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof comprises a light chain constant region selected from the group consisting of a kappa and a lambda light chain.

[0033] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, is optionally administered in combination with an additional therapeutic agent. The additional therapeutic agent may be a chemotherapeutic agent, an anti-PD-1 or PD-L1 antibody, an anti-CTLA4 antibody, a T-cell engager, a CD137-agonist-anti-FAP bispecific antibody, a tumor-targeting antibody, a VEGF-ANG2 bispecific antibody, a STING agonist, a MDM2 antagonist, or radiation therapy.

[0034] In one aspect, the anti-SIRP α antibody or an antigen-binding fragment thereof is administered in combination with an anti-PD-1 antibody, for example, nivolumab, pembrolizumab, pidilizumab, ezabenlimab, or atezolizumab. In a further aspect, the anti-SIRP α antibody or an antigen-binding fragment thereof is administered in combination with an anti-PD-L1 antibody including, for example, avelumab or durvalumab.

[0035] In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof is administered in combination with a tumor targeting antibody, for example an antibody that targets HER2 (e.g., trastuzumab), EGFR (e.g., cetuximab, panitumumab), CD20 (e.g., rituximab, ofatumumab), or CD52 (e.g., alemtuzumab).

[0036] In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof is administered in combination with two therapeutic agents. In a further aspect, the anti-SIRP α antibody or antigen-binding fragment thereof is administered in combination with an anti-PD-1 antibody (e.g., nivolumab, pembrolizumab, pidilizumab, ezabenlimab, or atezolizumab), or an anti-PD-L1 antibody (e.g., avelumab or durvalumab) and a tumor targeting antibody, for example and antibody that targets HER2 (e.g., trastuzumab), EGFR (e.g., tetuximab, panitumumab), CD20 (e.g., rituximab, ofatumumab), or CD52 (e.g., alemtuzumab).

[0037] In one aspect of the invention, the antibody or antigen-binding fragment thereof recognizes a specific linear or conformational "SIRP α epitope" on a SIRP α protein comprising the amino acid sequence set forth in any one of SEQ ID NOs: 240-252. In one aspect of the invention, the antibody or antigen-binding fragment thereof recognizes a specific linear or conformational "SIRP α epitope" on a SIRP α protein comprising the amino acid sequence set forth in any one of SEQ ID NOs: 253-260 and 264, in particular any one or more of SEQ ID NOS: 256 and 257. In one aspect of the invention the antibody or antigen-binding fragment thereof binds to a SIRP α

epitope comprising the amino acids: LEU 60, ILE 61, VAL 63, GLY 64, PRO 65, GLN 82, LYS 83, GLU 84, THR 97, LYS 98, ARG 99, GLU 100, LYS 126, GLY 127, SER 128, PRO 129 and ASP 130 as set forth in SEQ ID NO: 266. In one aspect of the invention the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising the amino acids LEU 60, ILE 61, VAL 63, GLY 64, PRO 65, GLN 82, LYS 83, GLU 84, THR 97, LYS 98, ARG 99, ASN 100, LYS 126, GLY 127, SER 128, PRO 129 and ASP 130 as set forth in SEQ ID NO: 265. In another aspect of the invention, the antibody or antigen-binding fragment thereof binds to the SIRP α epitope comprising the amino acid ASP130. In another aspect, the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising any one or more of SEQ ID NO: 253, SEQ ID NO: 254, SEQ ID NO: 255, SEQ ID NO: 264, and SEQ ID NO: 256, in particular SEQ ID NO: 256.

[0038] In one aspect of the invention the antibody or antigen-binding fragment thereof binds to the SIRP α epitope comprising the amino acids ARG 70, GLY 71, ALA 72, GLY 73, PRO 74, ALA 75, ARG 76, GLU 77, ALA 114, ALA 116, GLY 117, THR 118, TYR 120, THR 131, GLU 132, PHE 133, SER 135 and GLU 140 as set forth in SEQ ID NO: 266. In one aspect of the invention the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising the amino acids ARG 70, GLY 71, ALA 72, GLY 73, PRO 74, GLY 75, ARG 76, GLU 77, ALA 114, ALA 116, GLY 117, THR 118, TYR 120, VAL 132, GLU 133, PHE 134, SER 136 and GLU 141 as set forth in SEQ ID NO: 265. In another aspect of the invention, the antibody or antigen-binding fragment thereof binds to the SIRP α epitope comprising the amino acid ALA72. In another aspect, the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising any one or more of SEQ ID NO: 257, SEQ ID NO: 258, SEQ ID NO: 259 and SEQ ID NO: 260, in particular SEQ ID NO: 257. In another aspect of the invention, the antibody or antigen-binding fragment thereof blocks binding of any of Antibody A-E to SIRP α .

[0039] In another aspect, the antibody or antigen-binding fragment thereof exhibits comparable binding to (e.g., less than a 10-fold difference in affinity for) the modified SIRP α V1 polypeptide sequence of SEQ ID NO: 269 or 270, compared to its binding to the SIRP α V1 polypeptide of SEQ ID NO: 268, which optionally is measured at room temperature. Preferably the antibody or antigen-binding fragment thereof is antibody A, A4, A10, or an antigen-binding fragment thereof.

[0040] In another aspect, the antibody or antigen-binding fragment thereof exhibits reduced binding affinity for (e.g., at least a 10-fold reduction in affinity for) the modified SIRP α V1 polypeptide sequence of SEQ ID NO: 271 or 272, compared to its binding to the SIRP α V1

polypeptide of SEQ ID NO: 268, which optionally is measured at room temperature. Preferably the antibody or antigen-binding fragment thereof is antibody A, A4, A10, or an antigen-binding fragment thereof.

[0041] In another aspect, the antibody or antigen-binding fragment thereof exhibits comparable binding to (e.g., less than a 10-fold difference in affinity for) the modified SIRP α V2 polypeptide sequence of SEQ ID NO: 274 or 275, compared to its binding to the SIRP α V2 polypeptide of SEQ ID NO: 273, which optionally is measured at room temperature. Preferably the antibody or antigen-binding fragment thereof is antibody A, A4, A10, or an antigen-binding fragment thereof.

[0042] In another aspect, the antibody or antigen-binding fragment thereof exhibits reduced binding affinity for (e.g., at least a 10-fold reduction in affinity for) the modified SIRP β 1 polypeptide sequence of SEQ ID NO: 277, compared to its binding to the SIRP β 1 polypeptide of SEQ ID NO: 276, which optionally is measured at room temperature. Preferably the antibody or antigen-binding fragment thereof is antibody A, A4, A10, or an antigen-binding fragment thereof.

[0043] In another aspect, the antibody or antigen-binding fragment thereof exhibits binding to the modified polypeptide sequence of SEQ ID NO: 279, which optionally is measured at room temperature. Preferably the antibody or antigen-binding fragment thereof is antibody A, A4, A10, or an antigen-binding fragment thereof.

[0044] In another aspect, the antibody or antigen-binding fragment thereof exhibits binding to the modified polypeptide sequence of SEQ ID NO: 282 or 283, which optionally is measured at room temperature. Preferably the antibody or antigen-binding fragment thereof is antibody A, A4, A10, or an antigen-binding fragment thereof.

[0045] In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof exhibits comparable binding affinity for human SIRP α V1 and SIRP α V2, such as less than 10-fold difference (preferably less than 5-fold difference) in the binding affinity (K_D) for human SIRP α V1 and SIRP α V2, which optionally is measured at room temperature.

[0046] In one aspect, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof that competes for binding to V1-SIRP α and/or V2-SIRP α , with an anti-SIRP α antibody or antigen-binding fragment thereof of the invention. In one aspect, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof that competes for binding to V1-SIRP α and/or V2 SIRP α , with an antibody comprising a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 100, 110, 111, 112, 113, 114,

115, 116, or 117 and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 105, 125, or 126; or an antibody comprising a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, or 217 and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218.

[0047] In one aspect, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof that competes for binding to V1-SIRP α and/or V2-SIRP α , with an anti-SIRP α antibody or antigen-binding fragment thereof of the invention. In one aspect, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof that competes for binding to V1-SIRP α and/or V2 SIRP α , with an antibody comprising a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 104, 118, 119, 120, 121, 122, 123, 124, or 221 and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 109, 127, 128, 129, 130, or 222; or an antibody comprising a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 135, 153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, or 219 and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, or 220.

[0048] In one aspect, the invention provides a pharmaceutical composition comprising an anti-SIRP α antibody or antigen-binding fragment thereof as described above, and a pharmaceutically acceptable excipient, optionally in combination with one or more (e.g., one or two) additional therapeutic agents. In a further embodiment, the additional therapeutic agent is a chemotherapeutic agent, an anti-PD-1 or PD-L1 antibody, an anti-CTLA4 antibody, a T cell engager, a CD137-agonist-anti-FAP bispecific antibody, a tumor-targeting antibody, a VEGF-ANG2 bispecific antibody, a STING agonist, or a MDM2 antagonist.

[0049] In one aspect, the invention provides an anti-SIRP α antibody or antigen-binding fragment thereof as described above for use as a medicament or in the preparation of a medicament.

[0050] In one aspect, the invention provides a method of treating a SIRP α pathway disorder comprising administering to a subject in need thereof a pharmaceutically effective amount of the anti-SIRP α antibody or antigen-binding fragment thereof as described above. In one aspect, the invention provides an anti-SIRP α antibody or antigen-binding fragment thereof as described

above for use in treating a SIRP α pathway disorder. In one aspect, the invention provides the use of the anti- SIRP α antibody or antigen-binding fragment thereof as described above in manufacture of a medicament for treating a SIRP α pathway disorder.

[0051] In one aspect, the invention provides a method of modulating the interaction between SIRP α and CD47 in a subject (e.g., a human) comprising administering to the subject a composition comprising an anti-SIRP α antibody or the antigen-binding fragment as described above in an amount sufficient to block the CD47-mediated SIRP α signaling in the subject. In one embodiment, the invention provides an anti-SIRP α antibody or the antigen-binding fragment as described above for use in modulating the interaction between SIRP α and CD47 in a subject. In one embodiment, the invention provides the use of an anti-SIRP α antibody or the antigen-binding fragment as described above in the manufacture of a medicament for modulating the interaction between SIRP α and CD47 in a subject.

[0052] In one embodiment, the invention provides a method of enhancing phagocytosis comprising administering to a subject a composition comprising an anti-SIRP α antibody or the antigen-binding fragment as described above in an amount sufficient to block the CD47-mediated SIRP α signaling. In one embodiment, the invention provides an anti-SIRP α antibody or the antigen-binding fragment as described above for use in enhancing phagocytosis by macrophages and/or dendritic cells in a subject. In one embodiment, the invention provides an anti-SIRP α antibody or the antigen-binding fragment as described above for use in enhancing phagocytosis of tumor cells by macrophages and/or dendritic cells in a subject. In one embodiment, the present invention provides the use of an anti-SIRP α antibody or the antigen-binding fragment as described above in the manufacture of a medicament for a subject. In one aspect of the invention, the invention provides a method of enhancing antibody dependent cellular phagocytosis (ADCP), in combination with a tumor targeting agent, preferably a tumor targeting antibody, the method comprising administering to a subject a composition comprising an anti-SIRP α antibody or the antigen-binding fragment as described above in an amount sufficient to block the CD47-mediated SIRP α signaling in combination with a tumor targeting agent, preferably a tumor targeting antibody, more preferably a tumor targeting antibody that targets HER2 (e.g., trastuzumab), EGFR (e.g., cetuximab, panitumumab), CD20 (e.g., rituximab, ofatumumab) CD52 (e.g., alemtuzumab).

[0053] In one embodiment, in a method above, in an anti-SIRP α antibody or antigen-binding fragment thereof for use above, or in the use of an anti-SIRP α antibody or antigen-binding

fragment thereof above, the disease is selected from the group consisting of cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis.

[0054] In one embodiment, in the method above, in the anti-SIRP α antibody or antigen-binding fragment thereof for use above, or in the use of the anti-SIRP α antibody or antigen-binding fragment thereof above, the antibody or antigen-binding fragment thereof is administered by a parenteral route, intravenous route, or subcutaneous route of administration.

[0055] In one embodiment, the invention provides an isolated polynucleotide encoding a heavy chain variable region amino and/or a light chain variable region as described above.

[0056] In one embodiment, the invention provides an isolated polynucleotide encoding a heavy chain and/or a light chain as described above.

[0057] In one embodiment, the invention provides an isolated polynucleotide encoding a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 100, 101, 102, 103, 104, 105, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221.

[0058] In one embodiment, the invention provides an isolated polynucleotide encoding a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 105, 106, 107, 108, 109, 125, 126, 109, 127, 128, 129, 130, or 222.

[0059] In one embodiment, the invention provides an isolated polynucleotide encoding a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 100, 101, 102, 103, 104, 105, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221; and an isolated polynucleotide encoding a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 105, 106, 107, 108, 109, 125, 126, 109, 127, 128, 129, 130, or 222.

[0060] In one embodiment, the invention provides an isolated polynucleotide encoding a heavy chain region comprising the amino acid sequence of any one of SEQ NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, 217, 135, 153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 219, 133, 134, 137, 132, or 136.

[0061] In one embodiment, the invention provides an isolated polynucleotide encoding of a light chain region comprising the amino acid sequence of any one any one of SEQ NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218, 178, 196, 197, 198,

199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 220, 176, 177, 180, 175, or 179.

[0062] In one embodiment, the invention provides an isolated polynucleotide encoding a heavy chain region comprising the amino acid sequence of any one of SEQ NO: 131,138,139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, 217, 135,153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 219, 133, 134, 137, 132, or 136; and an isolated polynucleotide encoding of a light chain region comprising the amino acid sequence of any one any one of SEQ NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218, 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 220, 176, 177, 180, 175, or 179.

[0063] In one embodiment, the invention provides an expression vector comprising a polynucleotide as described above.

[0064] In one embodiment, the invention provides an expression vector comprising a polynucleotide encoding a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 100, 101, 102, 103, 104, 105, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221.

[0065] In one embodiment, the invention provides an expression vector comprising a polynucleotide encoding a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 105, 106, 107, 108, 109, 125, 126, 109, 127, 128, 129, 130, or 222.

[0066] In one embodiment, the invention provides an expression vector comprising a polynucleotide encoding a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 100, 101, 102, 103, 104, 105, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221; and a polynucleotide encoding a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 105, 106, 107, 108, 109, 125, 126, 109, 127, 128, 129, 130, or 222.

[0067] In one embodiment, the invention provides an expression vector comprising a polynucleotide encoding a heavy chain region comprising the amino acid sequence of any one of SEQ NO: 131,138,139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, 217, 135,153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 219, 133, 134, 137, 132, or 136.

[0068] In one embodiment, the invention provides an expression vector comprising a polynucleotide encoding of a light chain region comprising the amino acid sequence of any one

any one of SEQ NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218, 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 220, 176, 177, 180, 175, or 179.

[0069] In one embodiment, the invention provides an expression vector comprising a polynucleotide encoding a heavy chain region comprising the amino acid sequence of any one of SEQ NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, 217, 135, 153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 219, 133, 134, 137, 132, or 136; and a polynucleotide encoding of a light chain region comprising the amino acid sequence of any one any one of SEQ NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218, 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 220, 176, 177, 180, 175, or 179.

[0070] In one embodiment, the invention provides a host cell comprising an expression vector as described above. In one embodiment, the host cell is a mammalian cell.

[0071] In one embodiment, the invention provides a method of manufacturing an antibody or antigen-binding fragment thereof comprising the steps of:

- culturing a host cell comprising an expression vector comprising an isolated polynucleotide encoding a heavy chain variable region as described above and an expression vector comprising polynucleotide encoding light chain variable region as described above under conditions that allow formation of an antibody or antigen-binding fragment thereof comprising both the heavy and light chain variable region, and
- recovering said antibody or antigen-binding fragment thereof.

[0072] In one embodiment, the invention provides a method of manufacturing an antibody or antigen-binding fragment thereof comprising the steps of:

- culturing a host cell comprising an expression vector comprising an isolated polynucleotide encoding a heavy chain as described above and comprising a polynucleotide encoding light chain as described above under conditions that allow formation of an antibody or antigen-binding fragment thereof comprising both the heavy and light chain variable region; and
- recovering said antibody or antigen-binding fragment thereof.

[0073] In one embodiment, a method above further comprises the step of purifying the antibody or antigen-binding fragment thereof. In one embodiment, a method above further

comprises the step of formulating the antibody or antigen-binding fragment thereof into a pharmaceutical composition. Also provided herein are pharmaceutical formulations comprising an anti-SIRP α antibody or an antigen-binding fragment thereof as disclosed herein.

BRIEF DESCRIPTION OF THE DRAWINGS

[0074] The summary, as well as the following detailed description, is further understood when read in conjunction with the appended drawings. For the purpose of illustrating the disclosed compositions, methods, and kits there are shown in the drawings exemplary embodiments of methods and kits; however, these should not be limited to the specific embodiments disclosed.

[0075] Figures 1A-1C are series of graphs depicting the binding of antibodies to full-length human V1-SIRP α or V2-SIRP α expressed on CHO cells. Figure 1A. Antibody binding to parental CHO cells. Figure 1B. Antibody binding to CHO cells expressing full-length human SIRP α V1 (NP_542970.1). Figure 1C. Antibody binding to CHO cells expressing full-length human SIRP α V2-expressing CHO cells (CAA71403.1).

[0076] Figures 2A-2B are series of graphs depicting the binding of antibodies to endogenous human V1-SIRP α or V2-SIRP α expressed on human monocytic cell lines. Figure 2A. Antibody binding to U-937 human monocytic cell line. Figure 2B. Antibody binding to THP-1 human monocytic cell line.

[0077] Figures 3A-3D are series of graphs depicting the binding of antibodies to endogenous human V1-SIRP α and/or V2-SIRP α expressed on primary human monocytes. Antibody binding to primary human CD14⁺ monocytes from donors homozygous for V1-SIRP α allele (Figures 3A-3D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0078] Figures 4A-4D are series of graphs depicting the binding of antibodies to endogenous human V1-SIRP α and/or V2-SIRP α expressed on primary human monocytes. Antibody binding to primary human CD14⁺ monocytes from donors heterozygous for V1- and V2-SIRP α alleles (Figures 4A-4D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0079] Figures 5A-5D are series of graphs depicting the binding of antibodies to endogenous human V1-SIRP α and/or V2-SIRP α expressed on primary human monocytes.

Antibody binding to primary human CD14⁺ monocytes from donors homozygous for V2-SIRP α allele (Figures 5A-5D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0080] Figures 6A-6D are series of graphs depicting the binding of antibodies to full-length human SIRP β 1 expressed on U973^{SIRP α KO} cells. Antibody binding to U937^{SIRP α KO} cells expressing full-length human SIRP β 1 (NP_006056.2) (Figures 6A-6D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0081] Figures 7A-7D are series of graphs depicting the binding of antibodies to full-length human SIRP β L expressed on U973^{SIRP α KO} cells. Antibody binding to U937^{SIRP α KO} cells expressing full-length human SIRP β L (NP_001129316.1) (Figures 7A-7D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0082] Figure 8 is a graph depicting the binding of antibodies to recombinant SIRP γ protein. Antibody binding to immobilized recombinant His-tagged human SIRP γ extracellular domain (NP_061026.2).

[0083] Figure 9 is a graph depicting the binding of antibodies to full-length human SIRP γ expressed on CHO cells. Antibody binding to CHO cells expressing full-length human SIRP γ (NP_061026.2).

[0084] Figures 10A-10D are series of graphs depicting the binding of antibodies to primary human CD3⁺ T cells. Antibody binding to primary human CD3⁺ T cells (Figures 10A-10D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0085] Figures 11A-11B are series of graphs depicting the blocking of human CD47 binding to human SIRP α . A. Antibodies block human CD47 binding to CHO cells expressing full-length human SIRP α V1 (NP_542970.1). B. Antibodies block human CD47 binding to CHO cells expressing full-length human SIRP α V2 (CAA71403.1).

[0086] Figures 12A-12D are series of graphs depicting the blocking of human CD47 binding to SIRP α expressed on primary cells. Antibodies block human CD47 binding to primary human CD14⁺ monocytes from donors homozygous for V1 (Figures 12A-12D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0087] Figures 13A-13D are series of graphs depicting the blocking of human CD47 binding to SIRP α expressed on primary cells. Antibodies block human CD47 binding to primary human CD14⁺ monocytes from donors heterozygous for V1- and V2-SIRP α alleles. Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0088] Figures 14A-14D are series of graphs depicting the blocking of human CD47 binding to SIRP α expressed on primary cells. Antibodies block human CD47 binding to primary human CD14⁺ monocytes from donors homozygous for V2 alleles. Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0089] Figures 15A-15D are series of graphs showing that the cellular CD47-mediated SIRP α activation blocked by anti-SIRP α antibodies. Antibodies dose-dependently block cellular CD47-mediated V1-SIRP α signaling (Figures 15A-15D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0090] Figures 16A-16D are series of graphs showing that the cellular CD47-mediated SIRP α activation blocked by anti-SIRP α antibodies. Antibodies dose-dependently block cellular CD47-mediated V2-SIRP α signaling (Figures 16A-16D). Solid, dashed, and dotted lines indicate antibody molecules engineered on hIgG1 (LALA), hIgG4P, hIgG1 (K322A) backbones, respectively.

[0091] Figures 17A-17B are series of histograms showing that CD47-mediated inhibition of phagocytosis by human monocytic cell lines can be restored by anti-SIRP α antibodies. Anti-SIRP α antibodies restore (Figure 17A) V1- and (Figure 17B) V2-SIRP α expressing U937^{SIRP α KO} cell's ability to phagocytize CD47-coated beads.

[0092] Figures 18A-18C are series of histograms and a graph demonstrating that anti-SIRP α antibodies restore CD47-mediated inhibition of phagocytosis by primary human macrophages derived from donors homozygous for V1-SIRP α allele. Figure 18A. SIRP α antagonists restore monocyte-derived macrophage ability to phagocytize opsonized CD47-coated beads. Figure 18B. SIRP α antagonists enhance monocyte-derived macrophage ability to phagocytize non-opsonized CD47-coated beads. Figure 18C. SIRP α antagonists dose-dependently restores monocyte-derived macrophage ability to phagocytize opsonized CD47-coated beads.

[0093] Figures 19A-19C are series of histograms and a graph demonstrating that anti-SIRP α antibodies restore CD47-mediated inhibition of phagocytosis by primary human macrophages derived from donors heterozygous for V1- and V2-SIRP α allele. Figure 19A. SIRP α antagonists restore monocyte-derived macrophage ability to phagocytize opsonized CD47-coated beads. Figure 19B. SIRP α antagonists restore monocyte-derived macrophage ability to phagocytize non-opsonized CD47-coated beads. Figure 19C. SIRP α antagonists dose-dependently restores monocyte-derived macrophage ability to phagocytize opsonized CD47-coated beads.

[0094] Figures 20A-20C are series of histograms and a graph demonstrating that anti-SIRP α antibodies restore CD47-mediated inhibition of phagocytosis by primary human macrophages derived from donors homozygous for V2-SIRP α allele. Figure 20A. SIRP α antagonists restore monocyte-derived macrophage ability to phagocytize opsonized CD47-coated beads. Figure 20B. SIRP α antagonists restore monocyte-derived macrophage ability to phagocytize non-opsonized CD47-coated beads. Figure 20C. SIRP α antagonists dose-dependently restores monocyte-derived macrophage ability to phagocytize opsonized CD47-coated beads.

[0095] Figures 21A-21B are series of histograms showing that anti-SIRP α antibodies restore CD47-mediated inhibition of phagocytosis by primary human dendritic cells derived from donors homozygous for V1-SIRP α allele. Figure 21A. SIRP α antagonists restore monocyte-derived dendritic cells' ability to phagocytize opsonized CD47-coated beads. Figure 21B. SIRP α antagonists restore monocyte-derived dendritic cells' ability to phagocytize non-opsonized CD47-coated beads.

[0096] Figures 22A-22B are series of histograms showing that anti-SIRP α antibodies restore CD47-mediated inhibition of phagocytosis by primary human dendritic cells derived from donors heterozygous for V1- and V2-SIRP α allele. Figure 22A. SIRP α antagonists restore monocyte-derived dendritic cells' ability to phagocytize opsonized CD47-coated beads. Figure 22B. SIRP α antagonists restore monocyte-derived dendritic cells' ability to phagocytize non-opsonized CD47-coated beads.

[0097] Figures 23A-23B are series of histograms showing that anti-SIRP α antibodies restore CD47-mediated inhibition of phagocytosis by primary human dendritic cells derived from donors homozygous for V2-SIRP α allele. Figure 23A. SIRP α antagonists restore monocyte-derived dendritic cells' ability to phagocytize opsonized CD47-coated beads. Figure 23B. SIRP α

antagonists restore monocyte-derived dendritic cells' ability to phagocytize non-opsonized CD47-coated beads.

[0098] Figures 24A-24B are series of histograms showing that anti-SIRP α antibodies restore human CD47-mediated inhibition of MLR. Human CD47-Fc mediated immunosuppression of MLR partially restored by anti-SIRP α mAb alone or in combination with anti-PD1 antagonist, pembrolizumab (Figures 24A-24B).

[0099] Figures 25A-25F are series of images depicting the epitopes recognized by anti-SIRP α antibodies. Structures of Fab fragments of Antibody E (Figure 25A) or Antibody A (Figure 25B) bound to domain 1 of the extracellular domain of human SIRP α -V2 (residues 1 to 115). Figure 25C. Structure of CD47 (in dark grey) bound to domain 1 of the extracellular domain of human SIRP α -V2 (PDB: 2JJS). Figures 25D-25F: structure of domain 1 of the extracellular domain of human SIRP α -V2 highlighting the residues contacted by the Fab fragment of Antibody E (Figure 25D), by the Fab fragment of Antibody A (Figure 25E), or human CD47 (Figure 25F).

[00100] Figures 26A-26B are series of images depicting epitopes of Antibody A::SIRP α _V2 complex and Antibody E::SIRP α _V2 complex based on crystal structures. Figure 26A depicts SIRP α V2 shown in light grey as stick representation and residues of Antibody A shown in dark grey by atom type. SIRP α V1 residues comprising atoms in a distance < 4.5Å to Fab fragments of Antibody A are marked and extended by column to the sequences of SIRP α V1 and SIRP γ , aligned by sequence with the program MOE. The interaction of SIRP α V2 D130 is highlighted as a major difference to SIRP γ , where this residue is a glutamate. Figure 26B depicts SIRP α V2 shown in light grey as stick representation and residues and surface of Antibody E shown in dark grey by atom type. SIRP α V1 residues comprising atoms in a distance < 4.5Å to Fab fragments of Antibody E are marked and extended by column to the sequences of SIRP α V1 and SIRP γ aligned by sequence with the program MOE. The interaction of SIRP α V2 A72 is highlighted as a major difference to SIRP γ , where this residue is comprised of the larger amino acid valine.

[00101] Figures 27A-27B, are series of histograms showing that anti-SIRP α antibodies block binding of SE5A5 to V1- or V2-expressing U-937 cells. Blockade of anti-SIRP α mAb, SE5A5, binding to (Figure 27A) V1- or (Figure 27B) V2-SIRP α expressing U-937 cells by anti-SIRP α antibodies.

[00102] Figures 28A-28C are series of graphs depicting the binding of antibodies to full-length cyno SIRP α expressed on CHO cells. Figure 28A. Antibody binding to CHO cells expressing full-length cyno SIRP α (EGM-02252, clone H3A9). Figure 28B. Antibody binding to

CHO cells expressing full-length cyno SIRP α (XP_015313155, clone P3HD10). Figure 28C. Antibody binding to CHO cells expressing full-length cyno SIRP α (NP_001271679, clone HC6).

[00103] Figures 29A-29B are series of graphs depicting the binding of antibodies to full-length cyno SIRP β 1 expressed on CHO cells. Figure 29A. Antibody binding to CHO cells expressing full-length cyno SIRP β 1 (XP_005568598, clone PA2). Figure 29B. Antibody binding to CHO cells expressing full-length cyno SIRP β 1v3 (XP_005568593, clone 1HC6).

[00104] Figure 30. Mean (SD) serum concentration-time profiles of Antibody A10 following IV administration in cynomolgus monkey at 5 mg/kg (filled squares) or 1 mg/kg (filled circles).

[00105] Figures 31A-31F show binding of antibodies to full-length human V1-SIRP α with various amino acid point mutations expressed on Expi-CHO cells. Results are shown for cells expressing full-length wild-type human SIRP α V1 (Figure 31A), SIRP α V1 N $\rightarrow$ E (Figures 31B), SIRP α V1 D $\rightarrow$ E (Figures 31C), SIRP α V1 D $\rightarrow$ N (Figure 31D), SIRP α V1 DD $\rightarrow$ EN (Figure 31E), and parental CHO cells (Figure 31F).

[00106] Figures 32A-32C show binding of antibodies to full-length human V2-SIRP α with various amino acid point mutations expressed on Expi-CHO cells. Results are shown for cells expressing full-length wild-type human SIRP α V2 (Figure 32A), SIRP α V2 E $\rightarrow$ N (Figure 32B), and SIRP α V2 D $\rightarrow$ E (Figure 32C).

[00107] Figures 33A-33B show binding of antibodies to full-length human SIRP β 1 with an amino acid point mutation expressed on Expi-CHO cells. Results are shown for cells expressing full length wild-type human SIRP β 1 (Figure 33A) and SIRP β 1 D $\rightarrow$ H (Figure 33B).

[00108] Figures 34A-34B show of antibodies to full-length human SIRP β L with an amino acid point mutation expressed on Expi-CHO cells. Results are shown for cells expressing wild-type SIRP β L (Figure 34A) or SIRP β L H $\rightarrow$ D (Figure 34B).

[00109] Figures 35A-35D show binding of antibodies to full-length human SIRP γ with various amino acid point mutations expressed on Expi-CHO cells. Results are shown for cells expressing full-length wild-type human SIRP γ (Figure 35A), SIRP γ E $\rightarrow$ D (Figure 35B), SIRP γ N $\rightarrow$ D (Figure 35C), and SIRP γ EN $\rightarrow$ DD (Figure 35D).

[00110] Figure 36A. Loops of SIRP α V1 (light grey, as observed in pdbID: 4CMM), SIRP α V2 (grey, as observed in the crystal structure in complex with Antibody A) both as shown as a schematic, and Antibody A (surface in dark grey, as observed in the crystal structure in complex with SIRP α V1).

[00111] Figure 36B. Loops of SIRP α V1 (light grey, as observed in pdbID: 4CMM), SIRP α V2 (grey, as observed in the crystal structure in complex with CD47 pdbID: 2JJS) both as shown as a schematic, and CD47 (surface in dark grey, as observed in the crystal structure in complex with SIRP α V1).

[00112] Figures 37A-37F. Loops of SIRP variants (light grey, color coded by atom type): conformation observed for (A) SIRP α V1 (pdbID: 4CMM), with numbering according to uniprot ID: P78324. (B) SIRP α V2 (as observed in the crystal structure in complex with Antibody A). (C) SIRP β 1 (pdbID: 2JJU). (D) SIRP β L (pdbID: 2JJV). (E) SIRP γ (pdbID: 2JJW). (F) Comparison of sequence motifs in the respective loop, with amino acid differences from SIRP α V1 that correlate with significant loss in binding being DD->EN in case of SIRP γ and D-> H in case of SIRP β L.

DETAILED DESCRIPTION OF THE INVENTION

[00113] This invention relates to anti-SIRP α antibodies or antigen-binding fragments thereof. The present invention addresses the need for treatments of conditions modulated by the CD47-mediated SIRP α signaling. In one aspect, the anti-SIRP α antibodies or antigen-binding fragments thereof of the invention are for diagnostic and/or therapeutic use, for example in a subject in need thereof such as a human.

[00114] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof specifically binds to SIRP α , in particular human or cynomolgus monkey SIRP α , more particularly human SIRP α . In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof binds to the V1 and/or V2 alleles of human SIRP α . In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof does not bind to SIRP γ , in particular cynomolgus monkey or human SIRP γ , more particularly human SIRP γ . In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, does not bind to rabbit, mouse, rat, or dog SIRP α .

[00115] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof has an EC₅₀ of 0.1 to 100 nM, 0.1 to 50 nM, 0.1 to 25 nM, 0.1 to 10 nM, or 0.1 to 5 nM. In a further aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof has an EC₅₀ of 0.1 nM, 0.2 nM, 0.3 nM, 0.4 nM, 0.5 nM, 0.6 nM, 0.7 nM, 0.8 nM, 0.9 nM, 1 nM, 2 nM, 3 nM, 4 nM, 5 nM, 6 nM, 7 nM, 8 nM, 9 nM, 10 nM, 11 nM, 12 nM, 13 nM, 14 nM, 15 nM, 16 nM, 17 nM, 18 nM, 19 nM, 20 nM, 21 nM, 22 nM, 23 nM, 24 nM, or 25 nM. The EC₅₀ may

be determined by any method known in the art including for example those set forth in the Examples.

[00116] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof has an IC₅₀ of 0.01 to 100 nM, 0.01 to 50 nM, 0.01 to 25 nM, 0.01 to 10 nM, or 0.01 to 5 nM. In a further aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof has an IC₅₀ of 0.01 nM, 0.02 nM, 0.03 nM, 0.04 nM, 0.05 nM, 0.06 nM, 0.07 nM, 0.08 nM, 0.09 nM, 0.1 nM, 0.2 nM, 0.3 nM, 0.4 nM, 0.5 nM, 0.6 nM, 0.7 nM, 0.8 nM, 0.9 nM, 1 nM, 2 nM, 3 nM, 4 nM, 5 nM, 6 nM, 7 nM, 8 nM, 9 nM, 10 nM, 11 nM, 12 nM, 13 nM, 14 nM, 15 nM, 16 nM, 17 nM, 18 nM, 19 nM, 20 nM, 21 nM, 22 nM, 23 nM, 24 nM, or 25 nM. The IC₅₀ may be determined by any method known in the art including for example those set forth in the Examples.

[00117] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, binds to human SIRP α -V1 and SIRP α -V2 at a high affinity. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, binds to human SIRP α -V1 (e.g., human SIRP α -V1 comprising the amino acid sequence set forth in SEQ ID NO:240) at a high affinity, for example at an affinity of 20 nM or less, for example 10 nM or less, for example 5 nM or less, for example 1 nM or less. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, binds to human SIRP α -V2 (e.g., human SIRP α -V2 comprising the amino acid sequence set forth in SEQ ID NO:241) at a high affinity, for example at an affinity of 20 nM or less, for example 10 nM or less, for example 5 nM or less, for example 1 nM or less. In another aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, binds to cynomolgus monkey SIRP α . In another aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof, binds to cynomolgus monkey SIRP α (e.g., cynomolgus monkey SIRP α comprising the amino acid sequence set forth in SEQ ID NO:247) at an affinity of 400 nM or less, 300 nM or less, 250 nM or less, 200 nM or less, 100 nM or less, or 50 nM or less. In another aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof binds to cynomolgus monkey SIRP α (e.g., cynomolgus monkey SIRP α comprising the amino acid sequence set forth in SEQ ID NO:248) at an affinity of 400 nM or less, 300 nM or less, 200 nM or less, or 50 nM or less. In another aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof binds to cynomolgus monkey SIRP α (e.g., cynomolgus monkey SIRP α comprising the amino acid sequence set forth in SEQ ID NO:249) at an affinity of 400 nM or less, 300 nM or less, 200 nM or less, or 50 nM or less.

[00118] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof does not bind to SIRP γ , for example at an affinity of 1 μ M or greater. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof does not bind to human SIRP γ , for example does not bind to human SIRP γ at an affinity of greater than 1 μ M or greater. In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof does not bind to cynomolgus monkey SIRP γ , for example does not bind to cynomolgus monkey SIRP γ at an affinity of 1 μ M or greater.

[00119] In one aspect, an anti-SIRP α antibody or antigen-binding fragment thereof of the invention blocks the interaction between SIRP α and CD47. In a further aspect, an anti-SIRP α antibody or antigen-binding fragment thereof of the invention blocks CD47-mediated SIRP α signaling. In some aspects, the antibody of the invention blocks the binding of CD47 to SIRP α , whereby it decreases CD47-mediated SIRP α signaling by at least 80%, by at least 85%, by at least 90%, or by at least 95% when compared with a comparator antibody control or in the absence of an anti-SIRP α antibody or antigen-binding fragment of the invention. In an embodiment, the comparator antibody control comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 100, 101, 102, 103, 104, 105, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 105, 106, 107, 108, 109, 125, 126, 109, 127, 128, 129, 130, or 222; or a heavy chain region comprising the amino acid sequence of any one of SEQ NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, 217, 135, 153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 219, 133, 134, 137, 132, or 136; and a light chain region comprising the amino acid sequence of any one any one of SEQ NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218, 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 220, 176, 177, 180, 175, or 179.

[00120] Whether a binding domain specifically binds to a target can be tested with various methods known in the art. These methods include, Surface Plasmon Resonance or ELISA to detect binding of antibodies to purified proteins, or flow cytometry to detect binding of antibodies to cells. The ability of an antibody to block binding of CD47 can be measured by detecting purified soluble CD47 interaction with SIRP α -expressing cells. Alternatively, the blocking activity of an antibody can be measured by assessing SIRP α phosphorylation and recruitment of SHP-1 phosphatase. The blocking activity of an anti-SIRP α antibody can also be

evaluated by the capacity to restore the inhibition of phagocytosis caused by CD47. Methods for determining antibody specificity and affinity by competitive inhibition are known in the art.

[00121] In one aspect, the present invention provides an anti-SIRP α antibody, in particular a monoclonal anti-SIRP α antibody, for example a human monoclonal anti-SIRP α antibody, or a full-length human monoclonal antibody.

[00122] In one aspect, an anti-SIRP α antibody or antigen-binding fragment thereof of the present invention has favorable pharmacokinetic properties. In one aspect, an anti-SIRP α antibody of the present invention has favorable biophysical properties, for example yield, quality, stability or solubility.

Antibodies

[00123] The generalized structure of antibodies or immunoglobulin is well known to those of skill in the art, these molecules are heterotetrametric glycoproteins, typically of about 150,000 daltons, composed of two identical light (L) chains and two identical heavy (H) chains. Each light chain is covalently linked to a heavy chain by one disulfide bond to form a heterodimer, and the heterotrimeric molecule is formed through a covalent disulfide linkage between the two identical heavy chains of the heterodimers. Although the light and heavy chains are linked together by one disulfide bond, the number of disulfide linkages between the two heavy chains varies by immunoglobulin isotype. Each heavy and light chain also has regularly spaced intrachain disulfide bridges. Each heavy chain has at the amino-terminus a variable domain (V_H = variable heavy chain), followed by three or four constant domains (C_{H1} , C_{H2} , C_{H3} , and C_{H4}), as well as a hinge region between C_{H1} and C_{H2} . Each light chain has two domains, an amino-terminal variable domain (V_L = variable light chain) and a carboxy-terminal constant domain (C_L). The V_L domain associates non-covalently with the V_H domain, whereas the C_L domain is commonly covalently linked to the C_{H1} domain via a disulfide bond. Particular amino acid residues are believed to form an interface between the light and heavy chain variable domains (Chothia et al., 1985, J. Mol. Biol. 186:651-663, Vargas-Madrado E, Paz-García E. J Mol Recognit. 2003;16(3):113–120). The variable domains are also referred herein as variable regions, and the constant domains as constant regions.

[00124] Certain domains within the variable domains differ extensively between different antibodies i.e., are "hypervariable." These hypervariable domains contain residues that are directly involved in the binding and specificity of each particular antibody for its specific antigenic determinant. Hypervariability, both in the light chain and the heavy chain variable domains, is concentrated in three segments known as complementarity determining regions

(CDRs) or hypervariable loops (HVLs). CDRs are defined by sequence comparison in Kabat et al., 1991, In: Sequences of Proteins of Immunological Interest, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md., whereas HVLs are structurally defined according to the three-dimensional structure of the variable domain, as described by Chothia and Lesk, 1987, J. Mol. Biol. 196: 901-917. Where these two methods result in slightly different identifications of a CDR, the structural definition is preferred. As defined by Kabat, CDR-L1 is positioned at about residues 24-34, CDR-L2, at about residues 50-56, and CDR-L3, at about residues 89-97 in the light chain variable domain; CDR-H1 is positioned at about residues 31-35, CDR-H2 at about residues 50-65, and CDR-H3 at about residues 95-102 in the heavy chain variable domain. IMGT and NORTH provide alternative definitions of the CDRs (see, Lefranc MP. Unique database numbering system for immunogenetic analysis. Immunol Today (1997) 18:509; and North B, Lehmann A, Dunbrack RLJ. A new clustering of antibody CDR loop conformations. J Mol Biol. (2011) 406:228–56). Additionally, CDRs may be defined per the Chemical Computing Group (CCG) numbering (Almagro et al., Proteins 2011; 79:3050-3066 and Maier et al, Proteins 2014; 82:1599-1610). The CDR1, CDR2, CDR3 of the heavy and light chains therefore define the unique and functional properties specific for a given antibody.

[00125] The three CDRs within each of the heavy and light chains are separated by framework regions (FR), which contain sequences that tend to be less variable. From the amino terminus to the carboxy terminus of the heavy and light chain variable domains, the FRs and CDRs are arranged in the order: FR1, CDR1, FR2, CDR2, FR3, CDR3, and FR4. The largely β -sheet configuration of the FRs brings the CDRs within each of the chains into close proximity to each other as well as to the CDRs from the other chain. The resulting conformation contributes to the antigen binding site (see Kabat et al., 1991, NIH Publ. No. 91-3242, Vol. I, pages 647-669), although not all CDR residues are necessarily directly involved in antigen binding. Those skilled in the art can routinely determine which residues comprise a particular CDR given the variable region amino acid sequence of the antibody. The CDR1, CDR2, CDR3 of the heavy and light chains therefore define the unique and functional properties specific for a given antibody.

[00126] FR residues and Ig constant domains are generally not directly involved in antigen binding but contribute to antigen binding and/or mediate antibody effector function. Some FR residues are thought to have a significant effect on antigen binding in at least three ways: by noncovalently binding directly to an epitope, by interacting with one or more CDR residues, and by affecting the interface between the heavy and light chains. The constant domains are not directly involved in antigen binding but mediate various Ig effector functions, such as participation

of the antibody in antibody-dependent cellular cytotoxicity (ADCC), complement-dependent cytotoxicity (CDC) and antibody-dependent cellular phagocytosis (ADCP).

[00127] The light chains of vertebrate immunoglobulins are assigned to one of two clearly distinct classes, kappa (κ) and lambda (λ), based on the amino acid sequence of the constant domain. By comparison, the heavy chains of mammalian immunoglobulins are assigned to one of five major classes, according to the sequence of the constant domains: IgA, IgD, IgE, IgG, and IgM. IgG and IgA are further divided into subclasses (isotypes), e.g., IgG₁, IgG₂, IgG₃, IgG₄, IgA₁, and IgA₂, respectively. The heavy chain constant domains that correspond to the different classes of immunoglobulins are called α , δ , ϵ , γ , and μ , respectively. The subunit structures and three-dimensional configurations of the classes of native immunoglobulins are well known.

Definitions

[00128] The terms, "antibody", and "anti-SIRP α antibody", are used herein interchangeably and encompass monoclonal antibodies (including full length monoclonal antibodies), multispecific antibodies (e.g., bispecific antibodies), antibodies with minor modifications such as N- or C-terminal truncations and antibody fragments such as variable domains and other portions of antibodies that exhibit a desired biological activity, e.g., SIRP α binding.

[00129] The term "monoclonal antibody" refers to an antibody obtained from a substantially homogenous population of antibody molecules, i.e., the individual antibodies comprising the population are identical except for possible well-known alterations such as removal of C-terminal lysine from the antibody heavy chain or post-translational modifications such as amino acid isomerization or deamidation, methionine oxidation or asparagine or glutamine deamidation that may be present. Monoclonal antibodies typically bind one antigenic epitope. A bispecific monoclonal antibody binds two distinct antigenic epitopes. A monoclonal antibody may be monospecific or multispecific such as bispecific, monovalent, bivalent or multivalent. It should be understood that monoclonal antibodies can be made by any technique or methodology known in the art; including e.g., the hybridoma method (Kohler et al., 1975, Nature 256:495), or recombinant DNA methods known in the art (see, e.g., U.S. Pat. No. 4,816,567), or methods of isolation of monoclonal recombinantly produced using phage antibody libraries, using techniques described in Clackson et al., 1991, Nature 352: 624-628, and Marks et al., 1991, J. Mol. Biol. 222: 581-597.

[00130] Chimeric antibodies consist of the heavy and light chain variable regions of an antibody from one species (e.g., a non-human mammal such as a mouse) and the heavy and light chain constant regions of another species (e.g., human) antibody and can be obtained by linking the DNA sequences encoding the variable regions of the antibody from the first species (e.g., mouse) to the DNA sequences for the constant regions of the antibody from the second (e.g. human) species and transforming a host with an expression vector containing the linked sequences to allow it to produce a chimeric antibody. Alternatively, the chimeric antibody also could be one in which one or more regions or domains of the heavy and/or light chain is identical with, homologous to, or a variant of the corresponding sequence in a monoclonal antibody from another immunoglobulin class or isotype, or from a consensus or germline sequence. Chimeric antibodies can include fragments of such antibodies, provided that the antibody fragment exhibits the desired biological activity of its parent antibody, for example binding to the same epitope (see, e.g., U.S. Pat. No. 4,816,567; and Morrison et al., 1984, Proc. Natl. Acad. Sci. USA 81: 6851-6855).

[00131] The terms "antibody fragment", "antigen-binding fragment", "anti-SIRP α antibody fragment", "anti-SIRP α antibody fragment", "engineered anti-SIRP α antibody fragment" refer to a portion of a full length anti-SIRP α antibody, in which a variable region or a functional capability is retained, for example, SIRP α binding. Examples of antibody fragments include, but are not limited to, a Fab, Fab', F(ab')₂, Fd, Fv, scFv and scFv-Fc fragment, a diabody, a linear antibody, a single-chain antibody, a minibody, a diabody formed from antibody fragments, and multispecific antibodies formed from antibody fragments.

[00132] Antibody fragments can be obtained for example by treating full-length antibodies treated with enzymes such as papain or pepsin to generate useful antibody fragments. Papain digestion is used to produce two identical antigen-binding antibody fragments called "Fab" fragments, each with a single antigen-binding site, and a residual "Fc" fragment. The Fab fragment also contains the constant domain of the light chain and the C_{H1} domain of the heavy chain. Pepsin treatment yields a F(ab')₂ fragment that has two antigen-binding sites and is still capable of cross-linking antigen.

[00133] Another example of antibody fragments according to the invention are Fab' fragments. Fab' fragments differ from Fab fragments by the presence of additional residues including one or more cysteines from the antibody hinge region at the C-terminus of the C_{H1} domain. F(ab')₂ antibody fragments are pairs of Fab' fragments linked by cysteine residues in the hinge region. Other chemical couplings of antibody fragments are also known.

[00134] A "Fv" fragment contains a complete antigen-recognition and binding site consisting of a dimer of one heavy and one light chain variable domain in tight, non-covalent association. In this configuration, the three CDRs of each variable domain interact to define an antigen-binding site on the surface of the V_H - V_L dimer. Collectively, the six CDRs confer antigen-binding specificity to the antibody.

[00135] Antibody fragments may also include "single-chain Fv" or "scFv" fragments. A "single-chain Fv" or "scFv" antibody fragment is a single chain Fv variant comprising the V_H and V_L domains of an antibody where the domains are present in a single polypeptide chain. The single chain Fv is capable of recognizing and binding antigen. The scFv polypeptide may optionally also contain a polypeptide linker positioned between the V_H and V_L domains in order to facilitate formation of a desired three-dimensional structure for antigen binding by the scFv (see, e.g., Pluckthun, 1994, In *The Pharmacology of monoclonal Antibodies*, Vol. 113, Rosenberg and Moore eds., Springer-Verlag, New York, pp. 269-315).

[00136] Antibody fragments may also form tandem Fd segments, which comprise a pair of tandem Fd segments (V_H - C_{H1} - V_H - C_{H1}) to form a pair of antigen binding regions. These "linear antibodies" can be bispecific or monospecific as described in, for example, Zapata et al. 1995, *Protein Eng.* 8(10):1057-1062.

[00137] The term "human antibody" as used herein includes antibodies or fragments thereof derived from human germline immunoglobulin sequences. The term "human antibody" is not intended to include antibodies in which CDR sequences derived from the germline of another (mammalian) species, such as a mouse, rat or rabbit, have been grafted onto human framework sequences. Thus, as used herein, the term "human antibody" refers to an antibody or fragment thereof in which every part of the protein (e.g., CDR, framework, CL, CH domains (e.g., CH1, CH2, CH3), hinge, VL, VH) is substantially non-immunogenic in humans, with only minor sequence changes or variations as further described herein below.

[00138] Technologies for creating such a "human antibody" have been described and include without being limiting phage display or use of transgenic animals (www.Ablexis.com/technology-alivamab.php; WO 90/05144; D. Marks, H.R. Hoogenboom, T.P. Bonnert, J. McCafferty, A.D. Griffiths and G. Winter (1991) "By-passing immunisation. Human antibodies from V-gene libraries displayed on phage." *J.Mol.Biol.*, 222, 581-597; Knappik et al., *J. Mol. Biol.* 296: 57-86, 2000; S. Carmen and L. Jermutus, "Concepts in antibody phage display". *Briefings in Functional Genomics and Proteomics* 2002 1(2):189-203; Lonberg N, Huszar D. "Human antibodies from transgenic mice". *Int Rev Immunol.* 1995;13(1):65-93.; Brüggemann M,

Taussig MJ. "Production of human antibody repertoires in transgenic mice". *Curr Opin Biotechnol.* 1997 Aug;8(4):455-8.).

[00139] Thus, a human antibody is distinct from e.g., a chimeric or humanized antibody. It is pointed out that a human antibody can be produced by a non-human animal or prokaryotic or eukaryotic cell that is capable of expressing functionally rearranged human immunoglobulin (e.g., heavy chain and/or light chain) genes.

[00140] In one aspect, an anti-SIRP α antibody of the invention is a humanized antibody or antibody fragment thereof. A humanized antibody or a humanized antibody fragment is a specific type of chimeric antibody which includes an immunoglobulin amino acid sequence variant, or fragment thereof, which is capable of binding to a predetermined antigen and which, comprises one or more FRs having substantially the amino acid sequence of a human immunoglobulin and one or more CDRs having substantially the amino acid sequence of a non-human immunoglobulin. This non-human amino acid sequence often referred to as an "import" sequence is typically taken from an "import" antibody domain, particularly a variable domain. In general, a humanized antibody includes at least the CDRs or HVLs of a non-human antibody, inserted between the FRs of a human heavy or light chain variable domain. Methods of humanization of antibodies are for example described by Almagro et al., (2008) *Frontiers in Bioscience* 13, 1619-1633, or in WO12092374 A2.

[00141] The chimeric, humanized or human antibodies or antigen-binding fragments thereof of the present invention may further be engineered. Such engineering includes without limitation the removal or exchange of undesired amino acids, for example to reduce immunogenicity in humans, or to avoid deamidation, undesirable charges or lipophilicity or non-specific binding. Such removal or exchange of undesired amino acids can, for example, be introduced by random or site-specific mutagenesis in vitro or by somatic mutation in vivo. Moreover, in connection with chimeric or humanized antibodies, it will be understood that certain mouse FR residues may be retained in an antibody or fragment thereof.

[00142] In one aspect, an anti-SIRP α antibody comprises substantially all of at least one, and typically two, variable domains (such as contained, for example, in Fab, Fab', F(ab')₂, Fabc, and Fv fragments). In another aspect, an anti-SIRP α antibody also includes at least a portion of an immunoglobulin Fc region, typically that of a human immunoglobulin. Ordinarily, the antibody will contain both the light chain as well as at least the variable domain of a heavy chain. The antibody also may include one or more of the C_{H1}, hinge, C_{H2}, C_{H3}, and/or C_{H4} regions of the heavy chain, as appropriate.

[00143] In one aspect, an anti-SIRP α antibody can be selected from any class of immunoglobulins, including IgM, IgG, IgD, IgA and IgE, and any isotype, including IgG₁, IgG₂, IgG₃, IgG₄, IgA₁ and IgA₂. An alternative anti-SIRP α antibody can comprise sequences from more than one immunoglobulin class or isotype, and selecting particular modified or unmodified constant domains to optimize desired effector functions is within the ordinary skill in the art.

[00144] For example, the Fc region of an antibody mediates its serum half-life and effector functions, such as complement-dependent cytotoxicity (CDC), antibody-dependent cellular cytotoxicity (ADCC) and antibody-dependent cell phagocytosis (ADCP). Fc engineering can be employed to optimize antibody properties suited to the pharmacology activity required of them. Where such cytotoxic activity is not desirable, such as targeting an immune cell in the treatment of cancer, the constant domain may be of isotype with reduced effector function, such as IgG₄, and/or be modified with known modifications that reduce effector function. Where such cytotoxic activity is desirable, such as for destruction of a targeted tumor cell, the constant domain may be of isotype with increased effector function and/or be modified with known modifications to increase effector function. Several mutations are known to either reduce or increase effector function. See, e.g., *"The future of antibodies as cancer drugs"* Janice M Reichert, Eugen Dhimolea, Drug Discov Today (2012) Sep;17(17-18):954-63 - PMID: 22561895, *"Antibody Drug Discovery"* (Volume 4 of Molecular medicine and medicinal chemistry) Clive R. Wood, World Scientific, 2012 ISBN 1848166281, 9781848166288; *"Fc γ R requirements leading to successful immunotherapy"* Immunol Rev. (2015) Nov;268(1):104-22 - PMID: 26497516.

[00145] In one aspect, the constant domain of an antibody of the present invention is IgG4Pro, which has one replacement mutation (Ser228Pro) that prevents Fab-arm exchanging. In another aspect, the constant domain of an antibody of the present invention is IgG1, which has two mutations in the constant region, Leu234Ala and Leu235Ala to reduce effector function.

[00146] The FRs and CDRs, or HVLs, of an engineered anti-SIRP α antibody or antigen-binding fragment thereof need not correspond precisely to the parental sequences. For example, a parental sequence may be altered (e.g., mutagenized) by substitution, insertion or deletion such that the resulting amino acid residue is no longer identical to the original residue in the corresponding position in either parental sequence but the antibody nevertheless retains the function of binding to SIRP α . Such alteration typically will not be extensive and will be conservative alterations. Usually, at least 75% of the engineered antibody residues will

correspond to those of the parental sequences, more often at least 90%, and most frequently greater than 95%, or greater than 98% or greater than 99%.

[00147] Immunoglobulin residues that affect the interface between heavy and light chain variable regions ("the V_L - V_H interface") are those that affect the proximity or orientation of the two chains with respect to one another. Certain residues that may be involved in interchain interactions include V_L residues 34, 36, 38, 44, 46, 87, 89, 91, 96, and 98 and V_H residues 35, 37, 39, 45, 47, 91, 93, 95, 100, and 103 (utilizing the numbering system set forth in Kabat et al., Sequences of Proteins of Immunological Interest (National Institutes of Health, Bethesda, Md., 1987)). U.S. Pat. No. 6,407,213 also discusses that residues such as V_L residues 43 and 85, and V_H residues 43 and 60 also may be involved in this interaction. While these residues are indicated for human IgG only, they are applicable across species. Important antibody residues that are reasonably expected to be involved in interchain interactions are selected for substitution into the consensus sequence.

[00148] The terms "consensus sequence" and "consensus antibody" refer to an amino acid sequence which comprises the most frequently occurring amino acid residue at each location in all immunoglobulins of any particular class, isotype, or subunit structure, e.g., a human immunoglobulin variable domain. The consensus sequence may be based on immunoglobulins of a particular species or of many species. A "consensus" sequence, structure, or antibody is understood to encompass a consensus human sequence as described in certain embodiments, and to refer to an amino acid sequence which comprises the most frequently occurring amino acid residues at each location in all human immunoglobulins of any particular class, isotype, or subunit structure. Thus, the consensus sequence contains an amino acid sequence having at each position an amino acid that is present in one or more known immunoglobulins, but which may not exactly duplicate the entire amino acid sequence of any single immunoglobulin. The variable region consensus sequence is not obtained from any naturally produced antibody or immunoglobulin. Kabat et al., 1991, Sequences of Proteins of Immunological Interest, 5th Ed. Public Health Service, National Institutes of Health, Bethesda, Md., and variants thereof. The FRs of heavy and light chain consensus sequences, and variants thereof, provide useful sequences for the preparation of human or humanized anti-SIRP α antibodies. See, for example, U.S. Pat. Nos. 6,037,454 and 6,054,297.

[00149] An "isolated" antibody is one that has been identified and separated and/or recovered from a component of its natural environment or from a cell culture from which it was expressed. An isolated antibody or antibody fragment may have one or more co- or post-

translational modifications that arise during production, purification, and/or storage of the antibody or antibody fragment. Contaminant components of the antibody's natural environment are those materials that may interfere with diagnostic or therapeutic uses of the antibody, and can be enzymes, hormones, or other proteinaceous or non-proteinaceous solutes. In one aspect, the antibody will be purified to at least greater than 95% isolation by weight of antibody, for example purified to at least greater than 95%, 96%, 97%, 98%, or 99%.

[00150] An isolated antibody includes an antibody *in situ* within recombinant cells in which it is produced, since at least one component of the antibody's natural environment will not be present. Ordinarily however, an isolated antibody will be prepared by at least one purification step in which the recombinant cellular material is removed.

[00151] "Multispecific" refers to a protein, such as an antibody, that specifically binds two or more distinct antigens or two or more distinct epitopes within the same antigen.

[00152] "Bispecific" refers to a protein, such as an antibody, that specifically binds two distinct antigens or two distinct epitopes within the same antigen.

[00153] In some embodiments, the antibody that specifically binds SIRP α or the antigen-binding fragment thereof of the invention is a bispecific antibody. In some embodiments, the antibody or the antigen-binding fragment thereof of the invention is a multispecific antibody. The monospecific antibodies that specifically bind SIRP α provided herein may be engineered into bispecific antibodies, which are also encompassed within the scope of the invention.

[00154] Full-length bispecific antibodies may be generated for example using Fab arm exchange (e.g., half-molecule exchange, exchanging one heavy chain-light chain pair) between two monospecific bivalent antibodies by introducing substitutions at the heavy chain CH3 interface in each half molecule to favor heterodimer formation of two antibody half molecules having distinct specificity either *in vitro* in cell-free environment or using co-expression. The Fab arm exchange reaction is the result of a disulfide-bond.

[00155] Bispecific antibodies may also be generated using designs such as the Triomab/Quadroma (Trion Pharma/Fresenius Biotech), Knob-in-Hole (Genentech), CrossMAbs (Roche) and the electrostatically-induced CH3 interaction (Chugai, Amgen, NovoNordisk, Oncomed), the LUZ-Y (Genentech), the Strand Exchange Engineered Domain body (SEEDbody) (EMD Serono), the Biclonic (Merus) and as DuoBody® Products (Genmab A/S).

[00156] As used herein, the terms "identical" or "percent identity," in the context of two or more nucleic acids or polypeptide sequences, refer to two or more sequences or subsequences that are the same or have a specified percentage of nucleotides or amino acid residues that are the same, when compared and aligned for maximum correspondence. To determine the percent

identity, the sequences are aligned for optimal comparison purposes (e.g., gaps can be introduced in the sequence of a first amino acid or nucleic acid sequence for optimal alignment with a second amino or nucleic acid sequence). The amino acid residues or nucleotides at corresponding amino acid positions or nucleotide positions are then compared. When a position in the first sequence is occupied by the same amino acid residue or nucleotide as the corresponding position in the second sequence, then the molecules are identical at that position. The percent identity between the two sequences is a function of the number of identical positions shared by the sequences (i.e., % identity=# of identical positions/total # of positions (e.g., overlapping positions)x100). In some embodiments, the two sequences that are compared are the same length after gaps are introduced within the sequences, as appropriate (e.g., excluding additional sequence extending beyond the sequences being compared). For example, when variable region sequences are compared, the leader and/or constant domain sequences are not considered. For sequence comparisons between two sequences, a "corresponding" CDR refers to a CDR in the same location in both sequences (e.g., CDR-H1 of each sequence).

[00157] The determination of percent identity or percent similarity between two sequences can be accomplished using a mathematical algorithm. A preferred, non-limiting example of a mathematical algorithm utilized for the comparison of two sequences is the algorithm of Karlin and Altschul, 1990, Proc. Natl. Acad. Sci. USA 87:2264-2268, modified as in Karlin and Altschul, 1993, Proc. Natl. Acad. Sci. USA 90:5873-5877. Such an algorithm is incorporated into the NBLAST and XBLAST programs of Altschul et al., 1990, J. Mol. Biol. 215:403-410. BLAST nucleotide searches can be performed with the NBLAST program, score=100, wordlength=12, to obtain nucleotide sequences homologous to a nucleic acid encoding a protein of interest. BLAST protein searches can be performed with the XBLAST program, score=50, wordlength=3, to obtain amino acid sequences homologous to protein of interest. To obtain gapped alignments for comparison purposes, Gapped BLAST can be utilized as described in Altschul et al., 1997, Nucleic Acids Res. 25:3389-3402. Alternatively, PSI-Blast can be used to perform an iterated search which detects distant relationships between molecules (Id.). When utilizing BLAST, Gapped BLAST, and PSI-Blast programs, the default parameters of the respective programs (e.g., XBLAST and NBLAST) can be used. Another preferred, non-limiting example of a mathematical algorithm utilized for the comparison of sequences is the algorithm of Myers and Miller, CABIOS (1989). Such an algorithm is incorporated into the ALIGN program (version 2.0) which is part of the GCG sequence alignment software package. When utilizing the ALIGN program for comparing amino acid sequences, a PAM120 weight residue table, a gap length penalty of 12, and a gap penalty of 4 can be used. Additional algorithms for sequence analysis

are known in the art and include ADVANCE and ADAM as described in Torellis and Robotti, 1994, *Comput. Appl. Biosci.* 10:3-5; and FASTA described in Pearson and Lipman, 1988, *Proc. Natl. Acad. Sci. USA* 85:2444-8. Within FASTA, ktup is a control option that sets the sensitivity and speed of the search. If ktup=2, similar regions in the two sequences being compared are found by looking at pairs of aligned residues; if ktup=1, single aligned amino acids are examined. ktup can be set to 2 or 1 for protein sequences, or from 1 to 6 for DNA sequences. The default if ktup is not specified is 2 for proteins and 6 for DNA. Alternatively, protein sequence alignment may be carried out using the CLUSTAL W algorithm, as described by Higgins et al., 1996, *Methods Enzymol.* 266:383-402.

[00158] A "chemotherapeutic agent" is a chemical compound useful in the treatment of cancer. Examples of such chemotherapeutic agents include alkylating agents such as thiotepa and cyclophosphamide; alkyl sulfonates such as busulfan, improsulfan, and piposulfan; aziridines such as benzodopa, carboquone, meturedopa, and uredopa; ethylenimines and methylamelamines including altretamine, triethylenemelamine, trietylenephosphoramidate, triethylenethiophosphoramidate, and trimethylolmelamine; acetogenins (especially bullatacin and bullatacinone); camptothecin (including the synthetic analogue topotecan); bryostatin; callystatin; CC-1065 (including its adozelesin, carzelesin, and bizelesin synthetic analogues); cryptophycins (particularly cryptophycin 1 and cryptophycin 8); dolastatin, auristatins, (including analogues monomethyl-auristatin E and monomethyl-auristatin F); duocarmycin (including the synthetic analogues, KW-2189 and CBI-TMI); eleutherobin; pancratistatin; sarcodictyin; spongistatin; nitrogen mustards such as chlorambucil, chlornaphazine, cholophosphamide, estramustine, ifosfamide, mechlorethamine, mechlorethamine oxide hydrochloride, melphalan, novembichin, phenesterine, prednimustine; trofosfamide, uracil mustard; nitrosureas such as carmustine, chlorozotocin, fotemustine, lomustine, nimustine, ranimustine; antibiotics such as the enediyne antibiotics (e.g., calicheamicin, especially calicheamicin gamma11 and calicheamicin phil1, see for example, Agnew, *Chem. Intl. Ed. Engl.*, 33:183-186; dynemicin, including dynemicin A; bisphosphonates, such as clodronate; esperamicin; as well as neocarzinostatin chromophore and related chromoprotein enediyne antibiotic chromomorphores), aclacinomysins, actinomycin, authramycin, azaserine, bleomycins, cactinomycin, carabacin, caminomycin, carzinophilin, chromomycins, dactinomycin, daunorubicin, detorubicin, 6-diazo-5-oxo-L-norleucine, doxorubicin (Adriamycin™) (including morpholino-doxorubicin, cyanomorpholino-doxorubicin, 2-pyrrolino-doxorubicin, and deoxydoxorubicin), epirubicin, esorubicin, idarubicin, marcellomycin, mitomycins such as mitomycin C, mycophenolic acid, nogalamycin, olivomycins, peplomycin, potfiromycin, puromycin, quelamycin, rodorubicin, streptonigrin, streptozocin, tubercidin,

ubenimex, zinostatin, zorubicin; anti-metabolites such as methotrexate and 5-fluorouracil (5-FU); folic acid analogues such as denopterin, methotrexate, pteropterin, trimetrexate; purine analogs such as fludarabine, 6-mercaptopurine, thiamiprine, thioguanine; pyrimidine analogs such as ancitabine, azacitidine, 6-azauridine, carmofur, cytarabine, dideoxyuridine, doxifluridine, enocitabine, floxuridine; androgens such as calusterone, dromostanolone propionate, epitio stanol, mepitio stanane, testolactone; anti-adrenals such as aminoglutethimide, mitotane, trilostane; folic acid replenisher such as frolinic acid; aceglatone; aldophosphamide glycoside; aminolevulinic acid; eniluracil; amsacrine; bestrabucil; bisantrene; edatraxate; defofamine; democolcine; diaziquone; elfomithine; elliptinium acetate; an epothilone; etoglucid; gallium nitrate; hydroxyurea; lentinan; lonidamine; maytansinoids such as maytansine and ansamitocins; mitoguazone, mitoxantrone; mopidamol; nitracrine; pentostatin; phenamet; pirarubicin; losoxantrone; podophyllinic acid; 2-ethylhydrazide; procarbazine; PSK®; razoxane; rhizoxin; sizofuran; spirogermanium; tenuazonic acid; triaziquone; 2,2',2''-trichlorotriethylamine; trichothecenes (especially T-2 toxin, verracurin A, roridin A and anguidine); urethan; vindesine; dacarbazine; mannomustine; mitabronitol; mitolactol; pipobroman; gacytosine; arabinoside ("Ara-C"); cyclophosphamide; thiotepa; taxoids, e.g., paclitaxel (TAXOL®, Bristol-Myers Squibb Oncology, Princeton, N.J.) and doxetaxel (TAXOTERE®, Rhone-Poulenc Rorer, Antony, France); chlorambucil; gemcitabine (Gemzar™); 6-thioguanine; mercaptopurine; methotrexate; platinum analogs such as cisplatin and carboplatin; vinblastine; platinum; etoposide (VP-16); ifosfamide; mitoxantrone; vincristine; vinorelbine Navelbine™; novantrone; teniposide; edatrexate; daunomycin; aminopterin; xeloda; ibandronate; CPT-11; topoisomerase inhibitor RFS 2000; difluoromethylornithine (DMFO); retinoids such as retinoic acid; capecitabine; and pharmaceutically acceptable salts, acids, or derivatives of any of the above. Also included in this definition are anti-hormonal agents that act to regulate or inhibit hormone action on tumors such as anti-estrogens and selective estrogen receptor modulators (SERMs), including, for example, tamoxifen (including Nolvadex™), raloxifene, droloxifene, 4-hydroxytamoxifen, trioxifene, keoxifene, LY117018, onapristone, and toremifene (Fareston™); aromatase inhibitors that inhibit the enzyme aromatase, which regulates estrogen production in the adrenal glands, such as, for example, 4(5)-imidazoles, aminoglutethimide, megestrol acetate (Megace™), exemestane, formestane, fadrozole, vorozole (Rivisor™), letrozole (Femara™), and anastrozole (Arimidex™); and anti-androgens such as flutamide, nilutamide, bicalutamide, leuprolide, and goserelin; and pharmaceutically acceptable salts, acids, or derivatives of any of the above. Any one or more of these agents may be conjugated to the human antibodies or antigen-binding fragments thereof of

the present invention to provide a useful therapeutic agent for the treatment of various diseases and/or disorders.

[00159] For diagnostic as well as therapeutic monitoring purposes, the antibodies or antigen-binding fragment thereof of the invention also may be conjugated to a label, either a label alone or a label and an additional second agent (prodrug, chemotherapeutic agent and the like). A label, as distinguished from the other second agents refers to an agent that is a detectable compound or composition and it may be conjugated directly or indirectly to an anti-SIRP α antibody or antigen-binding fragment thereof of the present invention. The label may itself be detectable (e.g., radioisotope labels or fluorescent labels) or, in the case of an enzymatic label, may catalyze chemical alteration of a substrate compound or composition that is detectable. Labeled anti-SIRP α antibodies or antigen-binding fragments thereof can be prepared and used in various applications including in vitro and in vivo diagnostics.

[00160] In various aspects of the present invention one or more domains of the anti-SIRP α antibodies or antigen-binding fragments thereof will be recombinantly expressed. Such recombinant expression may employ one or more control sequences, i.e., polynucleotide sequences necessary for expression of an operably linked coding sequence in a particular host organism. The control sequences suitable for use in prokaryotic cells include, for example, promoter, operator, and ribosome binding site sequences. Eukaryotic control sequences include, but are not limited to, promoters, polyadenylation signals, and enhancers. These control sequences can be utilized for expression and production of anti-SIRP α antibodies or antigen-binding fragments thereof in prokaryotic and eukaryotic host cells.

[00161] A nucleic acid sequence is "operably linked" when it is placed into a functional relationship with another nucleic acid sequence. For example, a nucleic acid presequence or secretory leader is operably linked to a nucleic acid encoding a polypeptide if it is expressed as a preprotein that participates in the secretion of the polypeptide; a promoter or enhancer is operably linked to a coding sequence if it affects the transcription of the sequence; or a ribosome binding site is operably linked to a coding sequence if it is positioned so as to facilitate translation. Generally, "operably linked" means that the DNA sequences being linked are contiguous, and, in the case of a secretory leader, contiguous and in reading frame. However, enhancers are optionally contiguous. Linking can be accomplished by ligation at convenient restriction sites. If such sites do not exist, synthetic oligonucleotide adaptors or linkers can be used.

[00162] As used herein, the expressions "cell", "cell line", and "cell culture" are used interchangeably and all such designations include the progeny thereof. Thus, "transformants" and "transformed cells" include the primary subject cell and cultures derived therefrom without regard for the number of transfers, which may for example have been transfected with one or more expression vectors encoding one or more amino acids sequences of an antibody or antigen-binding fragment thereof of the present invention.

[00163] The term "mammal" for purposes of treatment according to the invention refers to any animal classified as a mammal, including humans, domesticated and farm animals, and zoo, sports, or pet animals, such as dogs, horses, cats, cows, and the like. Preferably, the mammal is a human.

[00164] A "disorder", as used herein, is any condition that would benefit from treatment with an anti-SIRP α antibody or antigen-binding fragment thereof described herein. This includes chronic and acute disorders or diseases including those pathological conditions that predispose the mammal to the disorder in question. Non-limiting examples of disorders to be treated herein include inflammatory, angiogenic, autoimmune and immunologic disorders, respiratory disorders, cancer, hematological malignancies, benign and malignant tumors, leukemias and lymphoid malignancies.

[00165] The terms "cancer" and "cancerous" refer to or describe the physiological condition in mammals that is typically characterized by unregulated cell growth. Examples of cancer include, but are not limited to, carcinoma, lymphoma, blastoma, sarcoma, and leukemia.

[00166] As used herein, the term "SIRP α pathway disorder" or "SIRP α pathway disease" refers to a condition, which can be alleviated by modulating the interaction between SIRP α and CD47, in particular by inhibiting the SIRP α /CD47 signaling. A "SIRP α pathway disorder" or "SIRP α pathway disease" includes myeloid associated diseases where SIRP α is expressed. A "SIRP α pathway disorder" or "SIRP α pathway disease" also includes conditions characterized by reduced phagocytosis by macrophages and/or dendritic cells that express SIRP α increased immune response is desired.

[00167] Examples of SIRP α pathway disorders are cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis. Examples of cancers include hematological cancer (e.g. leukemia, lymphoma, myeloma, e.g. multiple myeloma), and a metastatic lesion. Further examples include solid tumor cancers. Examples of solid tumors include malignancies, e.g. sarcomas and carcinomas, e.g. adenocarcinomas of the various organ

systems, such as those affecting the lung, breast, ovarian, lymphoid, gastrointestinal (e.g. colon), anal, genitals and genitourinary tract (e.g. renal, urothelial, bladder cells, prostate), pharynx, CNS (e.g. brain, neural or glial cells), head and neck, skin (e.g. melanoma), and pancreas, as well as adenocarcinomas which include malignancies such as colon cancers, rectal cancer, renal-cell carcinoma, liver cancer, gastric cancers, non-small cell lung cancer, cancer of the small intestine and cancer of the esophagus. The cancer may be at an early, intermediate, late stage or metastatic cancer.

[00168] In some embodiments, the cancer is chosen from a lung cancer (e.g. NSCLC (e.g. a NSCLC with squamous and/or non-squamous histology, or a NSCLC adenocarcinoma)), a melanoma (e.g. an advanced melanoma), a renal cancer (e.g. a renal cell carcinoma), a liver cancer, hepatocellular carcinoma, a myeloma (e.g. a multiple myeloma), a prostate cancer, a breast cancer (e.g. a breast cancer that does not express one, two or all of estrogen receptor, progesterone receptor, or HER2/neu, e.g. a triple negative breast cancer), a colorectal cancer, a pancreatic cancer, a head and neck cancer (e.g. head and neck squamous cell carcinoma (HNSCC), anal cancer, gastro-esophageal cancer, thyroid cancer, cervical cancer, a lymphoproliferative disease (e.g. a post-transplant lymphoproliferative disease) or a hematological cancer, T-cell lymphoma, B-cell lymphoma, a non-Hodgkin lymphoma, or a leukemia (e.g. a myeloid leukemia or a lymphoid leukemia).

[00169] In some embodiments, the cancer is chosen from a carcinoma (e.g. advanced or metastatic carcinoma), melanoma or a lung carcinoma, e.g. a NSCLC.

[00170] In some embodiments, the cancer is chosen from a pancreatic cancer, prostate cancer, breast cancer, colorectal cancer, lung cancer, glioblastoma, renal cancer, preferably pancreatic cancer, prostate cancer, breast cancer, colorectal cancer or lung cancer.

[00171] In some embodiments, the cancer is pancreatic cancer, lung cancer, breast cancer, melanoma, colorectal cancer, ovarian cancer, gastric cancer, thyroid cancer, liver cancer or prostate cancer.

[00172] The terms "specifically binds" or "specific binding" in the context of a binding agent, e.g., an antibody or antigen-binding fragment thereof, refers to a binding agent that associates more frequently, more rapidly, with greater duration, with greater affinity, with greater avidity or with some combination of the above, to an antigen or an epitope within the antigen than with an unrelated antigen. In certain embodiments, an antibody or antigen-binding fragment thereof specifically binds to an antigen or epitope within an antigen with a K_D of about 0.1 mM or

less, preferably less than about 1 μ M. Because of the sequence identity between homologous proteins in different species, or variants of a protein within a single species, specific binding can include an antibody or antigen-binding fragment thereof that recognizes a protein in more than one species (e.g., human SIRP α and cyno SIRP α). It is understood that, in certain embodiments, an antibody or antigen-binding fragment thereof that specifically binds a first protein may or may not specifically bind a second protein. As such, "specific binding" does not necessarily require (although it can include) exclusive binding, i.e. binding to a single protein. Thus, an antibody or antigen-binding fragment thereof may, in certain embodiments, specifically bind more than one protein.

[00173] Methods for determining whether two molecules specifically bind a protein are described herein or a known in the art and include, for example, equilibrium dialysis, surface plasmon resonance, and the like. In one embodiment, specific binding is characterized by a K_D of about 1×10^{-7} M (100 nM) or less according to the affinity binding method described in the Examples section herein. In another embodiment, specific binding is characterized by a K_D of about 5×10^{-8} M (50 nM) or less according to the affinity binding method described in the Examples section herein. In another embodiment, specific binding is characterized by a K_D of about 1×10^{-8} M (10 nM) or less according to the affinity binding method described in the Examples section herein. In another embodiment, specific binding is characterized by a K_D of about 5×10^{-9} M (5 nM) or less according to the affinity binding method described in the Examples section herein.

[00174] The term "subcutaneous administration" refers to introduction of a drug, for example an anti-SIRP α antibody or antigen-binding fragment thereof of the invention, under the skin of a subject such as an animal or human patient, preferable within a pocket between the skin and underlying tissue, by relatively slow, sustained delivery from a drug receptacle. Pinching or drawing the skin up and away from underlying tissue may create the pocket.

[00175] The term "subcutaneous infusion" refers to introduction of a drug, for example an anti-SIRP α antibody or antigen-binding fragment thereof of the invention, under the skin of a subject, preferably within a pocket between the skin and underlying tissue, by relatively slow, sustained delivery from a drug receptacle for a period of time including, but not limited to, 30 minutes or less, or 90 minutes or less. Optionally, the infusion may be made by subcutaneous implantation of a drug delivery pump implanted under the skin of the subject, wherein the pump delivers a predetermined amount of drug for a predetermined period of time, such as 30 minutes, 90 minutes, or a time period spanning the length of the treatment regimen.

[00176] The term "subcutaneous bolus" refers to drug administration beneath the skin of a subject, where bolus drug delivery is less than approximately 15 minutes; in another aspect, less than 5 minutes, and in still another aspect, less than 60 seconds. In yet even another aspect, administration is within a pocket between the skin and underlying tissue, where the pocket may be created by pinching or drawing the skin up and away from underlying tissue. For example, "subcutaneous bolus" refers to the administration of an anti-SIRP α antibody or antigen-binding fragment thereof of the invention to a subject in less than approximately 15 minutes; in another aspect, less than 5 minutes, and in still another aspect, less than 60 seconds

[00177] The term "therapeutically effective amount" is used to refer to an amount of an anti-SIRP α antibody or antigen-binding fragment thereof that relieves or ameliorates one or more of the symptoms of the disorder being treated. In doing so, it is that amount that has a beneficial patient outcome. Efficacy can be measured in conventional ways, depending on the condition to be treated.

[00178] The terms "treatment" and "therapy" and the like, as used herein, are meant to include therapeutic as well as prophylactic, or suppressive measures for a disease or disorder leading to any clinically desirable or beneficial effect, including but not limited to alleviation or relief of one or more symptoms, regression, slowing or cessation of progression of the disease or disorder. Thus, for example, the term treatment includes the administration of an anti-SIRP α antibody or antigen-binding fragment thereof prior to or following the onset of a symptom of a disease or disorder thereby preventing or removing one or more signs of the disease or disorder. As another example, the term includes the administration of an anti-SIRP α antibody or antigen-binding fragment thereof after clinical manifestation of the disease to combat the symptoms of the disease. Further, administration of an anti-SIRP α antibody or antigen-binding fragment thereof after onset and after clinical symptoms have developed where administration affects clinical parameters of the disease or disorder, such as the degree of tissue injury or the amount or extent of metastasis, whether or not the treatment leads to amelioration of the disease, comprises "treatment" or "therapy" as used herein. Moreover, as long as the compositions of the invention either alone or in combination with another therapeutic agent alleviate or ameliorate at least one symptom of a disorder being treated as compared to that symptom in the absence of use of the anti-SIRP α antibody or antigen-binding fragment thereof composition or antigen-binding fragment thereof, the result should be considered an effective treatment of the underlying disorder regardless of whether all the symptoms of the disorder are alleviated or not.

[00179] The term "package insert" is used to refer to instructions customarily included in commercial packages of therapeutic products, that contain information about the indications, usage, administration, contraindications and/or warnings concerning the use of such therapeutic products.

Antibodies

[00180] Described and disclosed herein are anti-SIRP α antibodies, in particular human anti-SIRP α antibodies, as well as compositions and articles of manufacture comprising anti-SIRP α antibodies of the present invention. Also described are antigen-binding fragments of an anti-SIRP α antibody. The anti-SIRP α antibodies and antigen-binding fragments thereof can be used in the treatment of a variety of diseases or disorders, in particular diseases or disorders characterized by modulation of CD47-mediated SIRP α signaling. An anti-SIRP α antibody and an antigen-binding fragment thereof each include at least a portion that specifically recognizes a SIRP α epitope.

[00181] The epitopes are most commonly proteins, short oligopeptides, oligopeptide mimics (e.g., organic compounds that mimic antibody binding properties of the SIRP α antigen), or combinations thereof. The minimum size of a peptide or polypeptide epitope for an antibody is thought to be about four to five amino acids. Peptide or polypeptide epitopes contain for example at least seven amino acids or for example at least nine amino acids or for example between about 15 to about 20 amino acids. Since an antibody can recognize an antigenic peptide or polypeptide in its tertiary form, the amino acids comprising an epitope need not be contiguous, and in some cases, may not even be on the same peptide chain. Epitopes may be determined by various techniques known in the art, such as X-ray crystallography, Hydrogen/Deuterium Exchange Mass Spectrometry (HXMS), site-directed mutagenesis, alanine scanning mutagenesis, and peptide screening methods.

[00182] The generation of anti-SIRP α antibodies and their characterization is described in the Examples. CDRs of representative anti-SIRP α antibodies of the present invention are disclosed in Tables 1-25 below. Heavy Chain CDR-1, CDR-2, CDR3 (HCDR1-3) and Light Chain CDR-1, CDR-2, CDR3 (LCDR1-3) are provided according to the numbering systems according to Kabat, CCG, Chothia, IMGT, and North.

TABLE 1: KABAT NOMENCLATURE

	Antibody E, E1-E22		
	KABAT SEQ	SEQ ID NO:	
HCDR1	NYYWS	1	
HCDR1-h	DYYWS	223	
HCDR2	FIYYNGRTFYNNSSLKS	2	
HCDR2-h	FIYYNGRTFYNNPSLKS	3	
HCDR2-h	FIYDNGRTFYNNPSLKS	4	
HCDR2-h	FIYYTGRTFYNNPSLKS	5	
HCDR2-h	FIYYNGRTFYQSSLKS	224	
HCDR3	VRAYSGIGLDGTDV	6	
LCDR1	KSSQSLLYSNGYNYLD	7	
LCDR1-h	KSSQSLLYSNGYAYLD	8	
LCDR1-h	KSSQSLLYSNAYNYLD	9	
LCDR1-h	RSSQSLLYSTGYTYLD	225	
LCDR2	LGSNRAS	10	
LCDR2-h	QGSNRAS	11	
LCDR2-h	GGSSRAS	226	
LCDR3	MQALQTPLT	12	
LCDR3-h	GQALQTPLT	227	
HCDR1-Consensus	X1YYWS	228	wherein X1=N or D
HCDR2-Consensus	FIYX1X2GRTFYX3X4SLKS	229	wherein X1=Y or D; X2=N or T; X3=N or Q; X4=S or P
LCDR1-Consensus	X1SSQSLLYSX2X3YX4YLD	230	wherein X1=K or R; X2= N or T; X3=G or A; X4=N, A or T
LCDR2-Consensus	X1GSX2RAS	231	wherein X1=L, Q or G; X2=N or S
LCDR3-Consensus	X1QALQTPLT	232	wherein X1=M or G

TABLE 2: IMGT NOMENCLATURE

	Antibody E, E1-E22	
	IMGT SEQ	SEQ ID NO:
HCDR1	GGSI RNYY	13
HCDR2	IYYNGRT	14
HCDR2-h	IYDNGRT	15
HCDR2-h	IYYTGRT	16
HCDR3	ARVRAYSGIGLDGTDV	17
LCDR1	QSLLYSNGYNY	18

LCDR1-h	QSLLYSNGYAY	19
LCDR1-h	QSLLYSNAYNY	20
LCDR2	LGS	21
LCDR2-h	QGS	22
LCDR3	MQALQTPLT	12

TABLE 3: CCG NOMENCLATURE

	Antibody E, E1-E22	
	CCG SEQ	SEQ ID NO:
HCDR1	GGsirnyws	23
HCDR2	FIYYNGRTFYNSLKS	2
HCDR2-h	FIYYNGRTFYNSLKS	3
HCDR2-h	FIYDNGRTFYNSLKS	4
HCDR2-h	FIYYTGRTFYNSLKS	5
HCDR3	VRAYSGIGLDGTDV	6
LCDR1	KSSQSLLYSNGYNYLD	7
LCDR1-h	KSSQSLLYSNGYAYLD	8
LCDR1-h	KSSQSLLYSNAYNYLD	9
LCDR2	LGSNRAS	10
LCDR2-h	QGSNRAS	11
LCDR3	MQALQTPLT	12

TABLE 4: CHOTHIA NOMENCLATURE

	Antibody E, E1-E22	
	CHOTHIA SEQ	SEQ ID NO:
HCDR1	GGsirny	261
HCDR2	YYNGR	24
HCDR2-h	YDNGR	25
HCDR2-h	YYTGR	26
HCDR3	VRAYSGIGLDGTDV	6
LCDR1	KSSQSLLYSNGYNYLD	7
LCDR1-h	KSSQSLLYSNGYAYLD	8
LCDR1-h	KSSQSLLYSNAYNYLD	9
LCDR2	LGSNRAS	10
LCDR2-h	QGSNRAS	11
LCDR3	MQALQTPLT	12

TABLE 5: NORTH NOMENCLATURE

	Antibody E, E1-E22	
	NORTH SEQ	SEQ ID NO:
HCDR1	TVSGGSIRNYYS	27
HCDR2	FIYYNGRTF	28
HCDR2-h	FIYDNGRTF	29
HCDR2-h	FIYYTGRTF	30
HCDR3	ARVRAYSGIGLDGTDV	17
LCDR1	KSSQSLLYSNGYNYLD	7
LCDR1-h	KSSQSLLYSNGYAYLD	8
LCDR1-h	KSSQSLLYSNAYNYLD	9
LCDR2	YLGSNRAS	31
LCDR2-h	YQGSNRAS	32
LCDR3	MQALQTPLT	12

TABLE 6: KABAT NOMENCLATURE

	Antibody A, A1-A16		
	KABAT SEQ	SEQ ID NO:	
HCDR1	SYDMH	33	
HCDR2	AIGTAGDTYYTGSVKG	34	
HCDR3	GGVWDDAFDI	35	
LCDR1	RASQDINNYLA	36	
LCDR1-h	RASQGINNYAA	37	
LCDR2	TASSLHS	38	
LCDR3	QQYVSYPYT	39	
LCDR1-Consensus	RASQX1INNYX2A	233	wherein X1=D or G; X2=L or A

TABLE 7: IMGT NOMENCLATURE

	Antibody A, A1-A16	
	IMGT SEQ	SEQ ID NO:
HCDR1	GFTLSSYD	40
HCDR2	IGTAGDT	41
HCDR3	VRGGVWDDAFDI	42
LCDR1	QDINNY	43
LCDR1-h	QGINNY	44
LCDR2	TAS	45
LCDR3	QQYVSYPYT	39

TABLE 8: CCG NOMENCLATURE

Antibody A, A1-A16		
	CCG SEQ	SEQ ID NO:
HCDR1	GFTLSSYDMH	46
HCDR2	AIGTAGDTYYTGSVKG	34
HCDR3	GGVWDDAFDI	35
LCDR1	RASQDINNYLA	36
LCDR1-h	RASQGINNYAA	37
LCDR2	TASSLHS	38
LCDR3	QQVVSYPYT	39

TABLE 9: CHOTHIA NOMENCLATURE

Antibody A, A1-A16		
	CHOTHIA SEQ	SEQ ID NO:
HCDR1	GFTLSSY	47
HCDR2	GTAGD	48
HCDR3	GGVWDDAFDI	35
LCDR1	RASQDINNYLA	36
LCDR1-h	RASQGINNYAA	37
LCDR2	TASSLHS	38
LCDR3	QQVVSYPYT	39

TABLE 10: NORTH NOMENCLATURE

Antibody A, A1-A16		
	NORTH SEQ	SEQ ID NO:
HCDR1	AASGFTLSSYDMH	49
HCDR2	AIGTAGDTY	50
HCDR3	VRGGVWDDAFDI	42
LCDR1	RASQDINNYLA	36
LCDR1-h	RASQGINNYAA	37
LCDR2	YTASSLHS	51
LCDR3	QQVVSYPYT	39

TABLE 11: KABAT NOMENCLATURE

Antibody B		
	KABAT SEQ	SEQ ID NO:
HCDR1	GNYMH	52
HCDR2	WINPNSGGTNYAQKFQG	53
HCDR3	GSGWYENYYYYGMDV	54
LCDR1	RASQGISSWLA	55
LCDR2	AESSLQS	56
LCDR3	QQANSFPLT	57

TABLE 12: IMGT NOMENCLATURE

	Antibody B	
	IMGT SEQ	SEQ ID NO:
HCDR1	GYTFTGNY	58
HCDR2	INPNSGGT	59
HCDR3	VSGSGWYENYYYYGMDV	60
LCDR1	QGISSW	61
LCDR2	AES	62
LCDR3	QQANSFPLT	57

TABLE 13: CCG NOMENCLATURE

	Antibody B	
	CCG SEQ	SEQ ID NO:
HCDR1	GFTFSSYDMH	63
HCDR2	WINPNSGGTNYAQKFQG	53
HCDR3	GSGWYENYYYYGMDV	54
LCDR1	RASQGISSWLA	55
LCDR2	AESSLQS	56
LCDR3	QQANSFPLT	57

TABLE 14: CHOTHIA NOMENCLATURE

	Antibody B	
	CHOTHIA SEQ	SEQ ID NO:
HCDR1	GYTFTGN	64
HCDR2	NPNSGG	65
HCDR3	GSGWYENYYYYGMDV	54
LCDR1	RASQGISSWLA	55
LCDR2	AESSLQS	56
LCDR3	QQANSFPLT	57

TABLE 15: NORTH NOMENCLATURE

	Antibody B	
	NORTH SEQ	SEQ ID NO:
HCDR1	KASGYTFTGNMYH	66
HCDR2	WINPNSGGTN	67
HCDR3	VSGSGWYENYYYYGMDV	68
LCDR1	RASQGISSWLA	55
LCDR2	YAESSLQS	69
LCDR3	QQANSFPLT	57

TABLE 16: KABAT NOMENCLATURE

Antibody C		
	KABAT SEQ	SEQ ID NO:
HCDR1	SYDMH	33
HCDR2	VIGIAGDTYYPGSVKG	70
HCDR3	GGSWDDAFDI	71
LCDR1	RASQDINNYLA	36
LCDR2	TASSLQS	72
LCDR3	QQYVSYPYT	39

TABLE 17: IMGT NOMENCLATURE

Antibody C		
	IMGT SEQ	SEQ ID NO:
HCDR1	GFTFSSYD	73
HCDR2	IGIAGDT	74
HCDR3	ARGGSWDDAFDI	75
LCDR1	QDINNY	43
LCDR2	TAS	45
LCDR3	QQYVSYPYT	39

TABLE 18: CCG NOMENCLATURE

Antibody C		
	CCG SEQ	SEQ ID NO:
HCDR1	GFTFSSYDMH	76
HCDR2	VIGIAGDTYYPGSVKG	77
HCDR3	GGSWDDAFDI	78
LCDR1	RASQDINNYLA	36
LCDR2	TASSLQS	79
LCDR3	QQYVSYPYT	39

TABLE 19: CHOTHIA NOMENCLATURE

Antibody C		
	CHOTHIA SEQ	SEQ ID NO:
HCDR1	GFTFSSY	80
HCDR2	GIAGD	81
HCDR3	GGSWDDAFDI	82
LCDR1	RASQDINNYLA	36
LCDR2	TASSLQS	79
LCDR3	QQYVSYPYT	39

TABLE 20: NORTH NOMENCLATURE

Antibody C

	NORTH SEQ	SEQ ID NO:
HCDR1	AASGFTFSSYDMH	83
HCDR2	VIGIAGDTY	84
HCDR3	ARGGSWDDAFDI	85
LCDR1	RASQDINNYLA	36
LCDR2	YTASSLQS	86
LCDR3	QQYVSYPYT	39

TABLE 21: KABAT NOMENCLATURE

	Antibody D	
	KABAT SEQ	SEQ ID NO:
HCDR1	SFDMH	262
HCDR2	TIGIAGDTYFPGSVKG	87
HCDR3	GGNWDALDI	88
LCDR1	RASQDINNYLA	36
LCDR2	TASSLQS	72
LCDR3	QQYNTYPYT	89

TABLE 22: IMGT NOMENCLATURE

	Antibody D	
	IMGT SEQ	SEQ ID NO:
HCDR1	GFTFSSFD	263
HCDR2	IGIAGDT	74
HCDR3	ARGGNWDALDI	90
LCDR1	QDINNY	43
LCDR2	TAS	45
LCDR3	QQYNTYPYT	89

TABLE 23: CCG NOMENCLATURE

	Antibody D	
	CCG SEQ	SEQ ID NO:
HCDR1	GFTFSSFDMH	91
HCDR2	TIGIAGDTYFPGSVKG	92
HCDR3	GGNWDDALDI	93
LCDR1	RASQDINNYLA	36
LCDR2	TASSLQS	79
LCDR3	QQYNTYPYT	89

TABLE 24: CHOTHIA NOMENCLATURE

	Antibody D	
	CHOTHIA SEQ	SEQ ID NO:
HCDR1	GFTFSSF	94
HCDR2	GIAGD	81
HCDR3	GGNWDDALDI	95
LCDR1	RASQDINNYLA	36
LCDR2	TASSLQS	79
LCDR3	QQYNTYPYT	89

TABLE 25: NORTH NOMENCLATURE

	Antibody D	
	NORTH SEQ	SEQ ID NO:
HCDR1	AASGFTFSSFDMH	96
HCDR2	TIGIAGDTY	97
HCDR3	ARGGNWDDALDI	98
LCDR1	RASQDINNYLA	99
LCDR2	YTASSLQS	86
LCDR3	QQYNTYPYT	89

Anti- SIRP α Antibody Sequences

[00183] Heavy and light chain variable regions of representative anti-SIRP α antibodies of the present invention are disclosed in Tables 26-27 below.

TABLE 26: Heavy Chain Variable Region (VH) Amino Acid Sequences

>Antibody A
EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLEWWSAIGTAGDTYYTG SVKGRFTISRENAKNSLYLQMNSLRAGDTAVYYCVRGGVWDDAFDIWGQGTMTVTVSS (SEQ ID NO: 100)
>Antibody B
QVQLVQSGAEVKKPGASVKVSCASGYTFTGNYPMHWRQAPGQGLEWMGWINPNSGGTNY AQKFQGRVTMTTRDTSISTAYMELSRRLRSDDTAVYYCVSGSGWYENYYYYGMDVWGQGTTVT VSS (SEQ ID NO: 101)
>Antibody C
EVQLVESGGGLVQPGGSLRLSCAASGFTFSSYDMHWVRQATGKGLEWWSVIGIAGDTYYPGS VKGRFTISRENAKNSLYLQMNSLRVGDVAVYYCARGGSWDDAFDIWGQGTMTVTVSS (SEQ ID NO: 102)
>Antibody D
EVQLVESGGGLVQPGGSLRLSCAASGFTFSSFDHWRQPTGKGLEWWSVIGIAGDTYFPGS VKGRFTISRENAKNSLYLQMNSLRAGDTAVYYCARGGNWDDALDIWGQGTMTVTVSS (SEQ ID NO: 103)
>Antibody E
QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKLEWIGFIYYNGRTFYNSL KSRVTISLDMSMNQFSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQGTTVTVSSARFT VDKSSSTAYMQFSSLTSEDVAVYFCARSGPYSYYAGGYALDYWGQGTSTVTVSS (SEQ ID NO: 104)

TABLE 27: Light Chain Variable Region Amino Acid Sequences

>Antibody A
DIQMTQSPSSLSASVGDRVTITCRASQDINNYLAWFQQKPGKAPKSLIYTASSLHSGVPSKFSG SGSGTDFTLTISLQPEDFATYYCQQYVSYPTYFGGGTKLEIK (SEQ ID NO: 105)
>Antibody B
DIQMTQSPSSVSASVGDRVTITCRASQGISSWLAWYQQKPGKAPKLLIYAESSLQSGVPSRFS GSGSGTDFTLTISLQPEDFATYYCQQANSFPLTFGGGKVEIK (SEQ ID NO: 106)
>Antibody C
DIQMTQSPSSLSASIGDKVTITCRASQDINNYLAWFQQKPGKAPKSLIYTASSLQSGVPSKFSG SGSGTDFTLTISLQPEDFATYYCQQYVSYPTYFGGGTKLEIK (SEQ ID NO: 107)
>Antibody D
DIQMTQSPSSLSASVGDRVTITCRASQDINNYLAWFQQKPGKAPKSLIYTASSLQSGVPSKFSG SGSGTDFTLTISLQPEDFATYYCQQYNTYPPTYFGGGTKLEIK (SEQ ID NO: 108)
>Antibody E
DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYNYLDWYLQRPQGSPQLLIYLGSNRASGVP DRFSGSGSGTDFTLKISRVEAEDVGVFYCMQALQTPLTFGGGKVEIK (SEQ ID NO: 109)

[00184] Representative anti-SIRP α antibodies of the present invention have the light and/or heavy chain variable regions sequences as set forth in Tables 28 or 29.

TABLE 28: Heavy Chain Variable Region (VH) Amino Acid Sequences

CLONE NAME	VH SEQUENCE	SEQ ID NO:
Antibody A1 Antibody A9	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQAPGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRADDTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	110
Antibody A2 Antibody A10 Antibody A11	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQATGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRAADTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	111
Antibody A3 Antibody A12	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQATGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRASDTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	112
Antibody A4 Antibody A13	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQAPGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRAADTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	113
Antibody A5 Antibody A14	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQAPGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRASDTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	114
Antibody A6 Antibody A15	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQAPGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRAEDTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	115
Antibody A7	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWW RQATGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRAGDTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	116

Antibody A8	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWV RQAPGKGLEWWSAIGTAGDTYYTGSVKGRFTISRENAK NSLYLQMNSLRAGDTAVYYCVRGGVWDDAFDIWGQG TMVTVSS	117
Antibody A16		
Antibody E1	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWIGFIYYNGRTFYNPSLKSRVTISLDMSINQ FSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQ GTTVTVSS	118
Antibody E2		
Antibody E9		
Antibody E3	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWIGFIYYNGRTFYNPSLKSRVTISLDMSKN QFSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWG QGTTVTVSS	119
Antibody E4		
Antibody E12		
Antibody E5	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWIGFIYYNGRTFYNPSLKSRVTISLDKSKNQ FSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQ GTTVTVSS	120
Antibody E6		
Antibody E14		
Antibody E7	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWIGFIYYNGRTFYNPSLKSRVTISLDTSKNQ FSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQ GTTVTVSS	121
Antibody E10		
Antibody E16		
Antibody E8	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWMGFIYDNGRTFYNPSLKSRVTISLDMSM NQFSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVW GQGTTVTVSS	122
Antibody E11		
Antibody E17		

Antibody E13	QVQLQESGPGGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWIGFIYYTGRTFYNPSLKSRVTISLDMSINQ FSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQ GTTVTVSS	123
Antibody E15		
Antibody E20		
Antibody E18	QVQLQESGPGGLVRPSETLSLTCTVSGGSIRNYYWSWIR QPPGKGLEWIGFIYYTGRTFYNPSLKSRVTISLDMSKN QFSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQ QGTTVTVSS	124
Antibody E19		
Antibody E21		
Antibody E22	QVQLQESGPGGLVKPSETLSLTCTVSGGSIRDYYWSWIR QPPGKGLEWIGFIYYNGRTFYQSSLKSRVTISLDTSKNQ FSLKMTSVTAADTAVYYCARVRAYSGIGLDGTDVWGQ GTTVTVSS	221

TABLE 29: Light Chain Variable Region (VL) Amino Acid Sequences

CLONE NAME	VL SEQUENCE	SEQ ID NO:
Antibody A1	DIQMTQSPSSLSASVGDRVTITCRASQGGINNYAAWFQQ KPGKAPKSLIYTASSLHSGVPSKFSGSGSGTDFTLTIS LQPEDFATYYCQYVSYPTYFGQGTKLEIK	125
Antibody A2		
Antibody A3		
Antibody A4		
Antibody A5		
Antibody A6		
Antibody A7		
Antibody A8		

Antibody A9	DIQMTQSPSSLSASVGDRVTITCRASQDINNYLAWFQQ KPGKAPKSLIYTASSLHSGVPSKFSGSGSGTDFTLTSS LQPEDFATYYCQYVSYPTYTFGQGTKLEIK	126
Antibody A10		
Antibody A11		
Antibody A12		
Antibody A13		
Antibody A14		
Antibody A15		
Antibody A16		
Antibody E1	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLD WYLQRPQGQSPQLLIYQGSNRASGVDPDRFSGSGSGTDF TLKISRVEAEDVGVFYCMQALQTPLTFGGGTKVEIK	127
Antibody E3		
Antibody E5		
Antibody E7		
Antibody E8		
Antibody E13		
Antibody E19		
Antibody E2	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLD WYLQRPQGQSPQLLIYLGSNRASGVDPDRFSGSGSGTDF TLKISRVEAEDVGVYYCMQALQTPLTFGGGTKVEIK	128

Antibody E4	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLD WYLQRPQGQSPQLLIYLGSNRASGVPDRFSGSGSGTDF TLKISRVEAEDVGVIYCMQALQTPLTFGGGTKVEIK	129
Antibody E6		
Antibody E10		
Antibody E11		
Antibody E15		
Antibody E18		
Antibody E9	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNAYNYLD WYLQRPQGQSPQLLIYLGSNRASGVPDRFSGSGSGTDF TLKISRVEAEDVGVIYCMQALQTPLTFGGGTKVEIK	130
Antibody E12		
Antibody E14		
Antibody E16		
Antibody E17		
Antibody E20		
Antibody E21		
Antibody E22	DIVMTQSPLSLPVTPGEPASISCRSSQSLLYSTGYTYLD WYLQRPQGQSPQLLIYGGSSRASGVPDRFSGSGSGTDF TLKISRVEAEDVGVIYCGQALQTPLTFGQGTKVEIK	222

[00185] Representative anti-SIRP α antibodies of the present invention may comprise a heavy and/or light chain as set forth in Tables 30 or 31 below.

TABLE 30: FULL LENGTH HC SEQUENCES OF ANTI-SIRP α ANTIBODIES.

Antibody	Sequence	SEQ ID NO:
Antibody A	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAGDTAVY YCVRGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	131
Antibody F	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAGDTAVY YCVRGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPCSRSTSES TAALGCLVKDYFPEPTVSWNSGALTSGVHTFPAVLQSSGLYSLSSV TVPSSSLGTQTYTCNVDHKPSNTKVDKRVEPKYGPCCPPCPAPEFLGG PSVFLFPPKPKDTLMISRTPEVTCVVDVSDQEDPEVQFNWYVDGVE VHNAKTKPREEQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGLP SSIEKTISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIA VEWESNGQPENNYKTTTPVLDSDGSFFLYSRLTVDKSRWQEGNVFS CSVMHEALHNHYTQKSLSLSPG	132
Antibody B	QVQLVQSGAEVKKPGASVKVSCKASGYFTGNYMHWRQAPGQG LEWMGWINPNSGGTNYAQKFQGRVTMTTRDTSISTAYMELSRLSD DTAVYYCVSGSGWYENYYYYGMDVWGQGTITVTVSSASTKGPSVFP LAPSSKSTSGGTAALGCLVKDYFPEPTVSWNSGALTSGVHTFPAVL QSSGLYSLSSVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDK THTCPPCPAPEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSH DPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWL NGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQ VSLTCLVKGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKL TVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG	133

Antibody C	EVQLVESGGGLVQPGGSLRLSCAASGFTFSSYDMHWVRQATGKGLE WVSVIGIAGDTYYPGSVKGRFTISRENAKNSLYLQMNSLRVGD TAVY YCARGGSWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSGG TAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSVV TVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPAPEA AGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWYVD GVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNK ALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGFYPS DIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQQGN VFSCSVMH EALHNHYTQKSLSLSPG	134
Antibody E	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNSSLKSRVTISLDSMNQFSLKMTSVTAADTAVY YCARVRAYSIGILDGTDVWGQGT TVTVSSASTKGPSVFPLAPSSKST SGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSL SSVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCP APEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNW YVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV SNKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKG FYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMH EALHNHYTQKSLSLSPG	135
Antibody G	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNSSLKSRVTISLDSMNQFSLKMTSVTAADTAVY YCARVRAYSIGILDGTDVWGQGT TVTVSSASTKGPSVFPLAPCSRST SESTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSL SVTVPSSSLGTQTYTCNV DHKPSNTKVDKRVEPKSGPPCPAPEF LGGPSVFLFPPKPKDTLMISRTPEVTCVVDVVSQEDPEVQFNWYVD GVEVHNAKTKPREEQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNK GLPSSIEKTISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGFYPS DIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSRLTVDKSRWQEGN VFSCSVMH EALHNHYTQKSLSLSPG	136
Antibody D	EVQLVESGGGLVQPGGSLRLSCAASGFTFSSFDMHWVRQPTGKGLE WVSTIGIAGDTYFPGSVKGRFTISRENAKNSLYLQMNSLRAGDTAVY YCARGGNWDDALDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMH EALHNHYTQKSLSLSPG	137

Antibody A1	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRADDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	138
Antibody A2	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRADDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	139
Antibody A3	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRASDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	140
Antibody A4	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRADDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	141

Antibody A5	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRASDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	142
Antibody A6	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAEDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	143
Antibody A7	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAGDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	144
Antibody A8	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAGDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTWSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	145

Antibody A9	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRADDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSLKTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	146
Antibody A10	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRADDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSLKTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	147
Antibody A11	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRADDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPCSRSTSES TAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSSV VTPSSSLGTQTYICNVDHKPSNTKVDKRVEPKSGPPCPAPEFLGG PSVFLFPPKPKDTLMISRTPEVTCVVVDVSDQEDPEVQFNWYVDGVE VHNAKTKPREEQFNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKGLP SSIEKTISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIA VEWESNGQPENNYKTPPVLDSDGSFFLYSRLTVDKSRWQEGNVFS CSVMHEALHNHYTQKSLSLSLG	217
Antibody A12	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQATGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRASDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSLKTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	148

Antibody A13	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAADTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	149
Antibody A14	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRASDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	150
Antibody A15	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAEDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	151
Antibody A16	EVQLVESGGGLVQPGGSLRLSCAASGFTLSSYDMHWVRQAPGKGLE WVSAIGTAGDYYTGSVKGRFTISRENAKNSLYLQMNSLRAGDTAVY YCVRGGVWDDAFDIWGQGTMTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	152

Antibody E1	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDMSINQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	153
Antibody E2	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDMSINQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	154
Antibody E3	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDMSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	155
Antibody E4	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDMSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	156

Antibody E5	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDKSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGT TTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	157
Antibody E6	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDKSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGT TTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	158
Antibody E7	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDTSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGT TTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	159
Antibody E8	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WMGFIYDNGRTFYNP SLKSRVTISLDMSMNQFSLKMTSVTAADTAV YYCARVRAYSGIGLDGTDVWGQGT TTVSSASTKGPSVFPLAPSSKS TSGGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYS LSSVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPC PAPEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFN WYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKC KVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLV KGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSR WQQGNVFSCSVMHEALHNHYTQKSLSLSPG	160

Antibody E9	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDM SINQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	161
Antibody E10	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDT SKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	162
Antibody E11	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WMGFIYDNGRTFYNP SLKSRVTISLDM SMNQFSLKMTSVTAADTAV YYCARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKS TSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYS LSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPC PAPEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFN WYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKC KVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLV KGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSR WQQGNVFSCSVMHEALHNHYTQKSLSLSPG	163
Antibody E12	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDM SKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	164

Antibody E13	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYTGRTFYNP SLKSRVTISLDM SINQFSLKMTSVTAADTAVYYC ARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	165
Antibody E14	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	166
Antibody E15	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYTGRTFYNP SLKSRVTISLDM SINQFSLKMTSVTAADTAVYYC ARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	167
Antibody E16	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYNGRTFYNP SLKSRVTISLDSKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLSS SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	168

Antibody E17	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WMGFIYDNGRTFYNP SLKSRVTISLDM SMNQFSLKMTSVTAADTAV YYCARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKS TSGGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYS LSSVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPC PAPEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFN WYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCK KVSNAKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLV KGFYPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSR WQQGNVFSCSVMHEALHNHYTQKSLSLSPG	169
Antibody E18	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYTGRTFYNP SLKSRVTISLDM SKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	170
Antibody E19	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYTGRTFYNP SLKSRVTISLDM SKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	171
Antibody E20	QVQLQESGPGLVLPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYTGRTFYNP SLKSRVTISLDM SINQFSLKMTSVTAADTAVYYC ARVRAYSGIGLDGTDVWGQGTTTVSSASTKGPSVFPLAPSSKSTSG GTAALGCLVKDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS VVTVPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVS NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	172

Antibody E21	QVQLQESGPGLVRPSETLSLTCTVSGGSIRNYYWSWIRQPPGKGLE WIGFIYYTGRTFYNP SLKSRVTISLDM SKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGT TVVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSL SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	173
Antibody E22	QVQLQESGPGLVKRPSETLSLTCTVSGGSIRDYYWSWIRQPPGKGLE WIGFIYYNGRTFYQSSLSKSRVTISLDT SKNQFSLKMTSVTAADTAVYY CARVRAYSGIGLDGTDVWGQGT TVVSSASTKGPSVFPLAPSSKSTS GGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSL SVVTPSSSLGTQTYICNVNHKPSNTKVDKRVEPKSCDKTHTCPPCPA PEAAGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWY VDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKV NKALPAPIEKTISKAKGQPREPQVYTLPPSREEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSKLTVDKSRWQ QGNVFSCSVMHEALHNHYTQKSLSLSPG	219

TABLE 31: FULL LENGTH LC SEQUENCES OF ANTI-SIRP- α ANTIBODIES.

Antibody	Sequence	SEQ ID NO:
Antibody A	DIQMTQSPSSLSASVGDRVTITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSSPVTKSFNRGEC	174
Antibody F	DIQMTQSPSSLSASVGDRVTITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSSPVTKSFNRGEC	175
Antibody B	DIQMTQSPSSVSASVGDRVTITCRASQGISSWLAWYQQKPGKAPKL LIYAESSLQSGVPSRFSGSGSGTDFTLTISLQPEDFATYYCQQA NSFP LTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREA KVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKV YACEVTHQGLSSPVTKSFNRGEC	176
Antibody C	DIQMTQSPSSLSASIGDKVTITCRASQDINNYLAWFQQKPGKAPKSLI YTASSLQSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSSPVTKSFNRGEC	177

Antibody E	DIVMTQSPSLPVTGPGEPAISCKSSQSLYSNGYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPITFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	178
Antibody G	DIVMTQSPSLPVTGPGEPAISCKSSQSLYSNGYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPITFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	179
Antibody D	DIQMTQSPSSLSASVGDRVITICRASQDINNYLAWFQQKPGKAPKSL IYTASSLQSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYNTYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	180
Antibody A1	DIQMTQSPSSLSASVGDRVITICRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	181
Antibody A2	DIQMTQSPSSLSASVGDRVITICRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	182
Antibody A3	DIQMTQSPSSLSASVGDRVITICRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	183
Antibody A4	DIQMTQSPSSLSASVGDRVITICRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	184
Antibody A5	DIQMTQSPSSLSASVGDRVITICRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	185
Antibody A6	DIQMTQSPSSLSASVGDRVITICRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPY TFGGGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYKHKVY ACEVTHQGLSSPVTKSFNRGEC	186

Antibody A7	DIQMTQSPSSLSASVGDRVITITCRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPY TFGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVY ACEVTHQGLSPVTKSFNRGEC	187
Antibody A8	DIQMTQSPSSLSASVGDRVITITCRASQGINNYAAWFQQKPGKAPKS LIYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPY TFGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVY ACEVTHQGLSPVTKSFNRGEC	188
Antibody A9	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	189
Antibody A10	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	190
Antibody A11	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	218
Antibody A12	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	191
Antibody A13	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	192
Antibody A14	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	193
Antibody A15	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQQYVSYPYT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSPVTKSFNRGEC	194

Antibody A16	DIQMTQSPSSLSASVGDRVITITCRASQDINNYLAWFQQKPGKAPKSL IYTASSLHSGVPSKFSGSGSGTDFTLTISLQPEDFATYYCQYVSYPT FGQGTKLEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKV QWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADYEKHKVYA CEVTHQGLSSPVTKSFNRGEC	195
Antibody E1	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	196
Antibody E2	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVVYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	197
Antibody E3	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	198
Antibody E4	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVVYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	199
Antibody E5	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	200
Antibody E6	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVVYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	201
Antibody E7	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	202
Antibody E8	DIVMTQSPLSLPVTPGEPASISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVDPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPLTFGGGTKEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	203

Antibody E9	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	204
Antibody E10	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	205
Antibody E11	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	206
Antibody E12	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	207
Antibody E13	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	208
Antibody E14	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	209
Antibody E15	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	210
Antibody E16	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	211
Antibody E17	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPLTFGGGKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKDYSLSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	212

Antibody E18	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPITFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKSTYLSSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	213
Antibody E19	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNGYAYLDWYLQRPQGS PQLLIYQGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVFYCMQ ALQTPITFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKSTYLSSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	214
Antibody E20	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPITFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKSTYLSSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	215
Antibody E21	DIVMTQSPLSLPVTGPGEPAISCKSSQSLLYSNAYNYLDWYLQRPQGS PQLLIYLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQ ALQTPITFGGGTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNF YPREAKVQWKVDNALQSGNSQESVTEQDSKSTYLSSTLTLSKADY EKHKVYACEVTHQGLSSPVTKSFNRGEC	216
Antibody E22	DIVMTQSPLSLPVTGPGEPAISCRSSQSLLYSTGYTYLDWYLQRPQGS PQLLIYGGSSRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVFYCGQA LQTPITFGGQTKVEIKRTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFY PREAKVQWKVDNALQSGNSQESVTEQDSKSTYLSSTLTLSKADYE KHKVYACEVTHQGLSSPVTKSFNRGEC	220

[00186] Representative anti-SIRP α antibodies of the present invention may comprise a heavy and/or light chain constant region as set forth in Tables 32 or 33 below.

Table 32: EXAMPLE HC AND LC SEQUENCES OF THE CONSTANT REGIONS OF ANTI-SIRP α ANTIBODIES.

Antibody	Sequence	SEQ ID NO:
IgG1 - HC	ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTWNSGALTS GVHTFPAVLQSSGLYSLSSVTPVSSSLGTQTYICNVNHKPSNTKVKDK RVEPKSCDKHTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCV VVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTV LHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRE EMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTTPVLDSDG SFFLYSKLTVDKSRWQQGNVFCFSVMHEALHNHYTQKSLSLSPGK	234
IgG1 - LC	RTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL QSGNSQESVTEQDSKSTYLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC	235

IgG4 - HC	ASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVTVSWNSGALTS GVHTFPAVLQSSGLYSLSSVTVPSSSLGKTYTCNVDPKPSNTKVDK RVESKYGPPCPPCPAPEFLGGPSVFLFPPKPKDTLMISRTPEVTCVTV DVSQEDPEVQFNWYVDGVEVHNAKTKPREEQFNSTYRVVSVLTVLH QDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPREPQVYTLPPSQEE MTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSDGS FFLYSRLTVDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLGLK	236
IgG4 -LC	RTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL QSGNSQESVTEQDSKSTYLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC	237
IgG1-KO -HC	ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVTVSWNSGALTS GVHTFPAVLQSSGLYSLSSVTVPSSSLGKTQYICNVNHKPSNTKVDK RVEPKSCDKHTCPCPAPEAAAGGPSVFLFPPKPKDTLMISRTPEVTC VVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLT VLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPS REEMTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTPPVLDSD DGSFFLYSKLTVDKSRWQQGNVFCVMHEALHNHYTQKSLSLSPG K	238
IgG1-KO -LC	RTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAKVQWKVDNAL QSGNSQESVTEQDSKSTYLSSTLTLSKADYEKHKVYACEVTHQGLS SPVTKSFNRGEC	239

Amino Acid Sequence Variants

[00187] Variant anti-SIRP α antibodies and antibody fragments thereof can be engineered based on a set of CDRs depicted in Tables 1-25. It is to be understood that in the variant anti-SIRP α antibodies and antibody fragments the amino acid sequence of the CDRs remain unchanged or have minimal changes (e.g., 1-5 changes), but the surrounding regions, e.g., FR regions can be engineered. Amino acid sequence variants of the anti-SIRP α antibody can be prepared by introducing appropriate nucleotide changes into the anti-SIRP α antibody DNA, or by peptide synthesis. Such variants include, for example, deletions from, and/or insertions into and/or substitutions of, residues within the amino acid sequences of the anti-SIRP α antibodies of the examples herein. Any combination of deletions, insertions, and substitutions is made to arrive at the final construct, provided that the final construct possesses the desired characteristics. The amino acid changes also may alter post-translational processes of the human or variant anti-SIRP α antibody, such as changing the number or position of glycosylation sites.

[00188] In some embodiments, the present invention includes anti-SIRP α antibodies or antibody fragments thereof having a variable heavy chain and a variable light chain, wherein the variable heavy chain amino acid sequence and the variable light chain amino acid sequence are at least at least 90%, at least 92.5%, at least 95%, at least 98%, or at least 99% identical to

the amino acid sequences disclosed in Tables 26-29 provided that the antibody or fragments thereof retain binding to SIRP α -V1 and/or SIRP α -V2.

[00189] In some embodiments, the present invention includes anti-SIRP α antibodies or antibody fragments thereof having a variable heavy chain and a variable light chain, wherein the variable heavy chain amino acid sequence and the variable light chain amino acid sequence are at least 80%, at least 85%, at least 90%, at least 92.5%, at least 95%, at least 98%, or at least 99% identical to the amino acid sequences of SEQ ID Nos: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221, and SEQ ID Nos: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222, respectively.

[00190] In some embodiments, the present invention includes anti-SIRP α antibodies having a heavy chain and a light chain, wherein the heavy chain amino acid sequence and the light chain amino acid sequence are at least 95%, at least 98%, or at least 99% identical to the amino acid sequences disclosed in Tables 30 and 31 provided that the antibody or fragments thereof retain binding to SIRP α -V1 and/or SIRP α -V2.

[00191] In some embodiments, the anti-SIRP α antibodies or antibody fragments thereof comprise a variable heavy chain sequence that comprises an amino acid sequence with at least about 95%, about 96%, about 97%, about 98%, or about 99%, sequence identity to the amino acid sequence set forth in SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221. In other embodiments, the anti-SIRP α antibodies or antibody fragments thereof retains the binding and/or functional activity of an anti-SIRP α antibody or antibody fragment thereof that comprises the variable heavy chain sequence of SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221. In still further embodiments, the anti-SIRP α antibodies or antibody fragments thereof comprise the variable heavy chain sequence of SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221 and have one or more conservative amino acid substitutions, e.g., 1, 2, 3, 4, 5, 1-2, 1-3, 1-4 or 1-5 conservative amino acid substitutions in the heavy chain variable sequence. In yet further embodiments, the one or more conservative amino acid substitutions fall within one or more framework regions in SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221 (based on the numbering system of Kabat).

[00192] In some embodiments, the anti-SIRP α antibody or antibody fragment thereof comprises a variable heavy chain sequence with at least about 95%, about 96%, about 97%,

about 98%, or about 99% sequence identity to the anti-SIRP α heavy chain variable region sequence set forth in SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221 comprises one or more conservative amino acid substitutions in a framework region (based on the numbering system of Kabat), and retains the binding and/or functional activity of an anti-SIRP α antibody or antibody fragment thereof that comprises a variable heavy chain sequence as set forth in SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221 and a variable light chain sequence as set forth in SEQ ID NOs: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222.

[00193] In some embodiments, the anti-SIRP α antibodies or antibody fragments thereof comprise a variable light chain sequence that comprises an amino acid sequence with at least about 95%, about 96%, about 97%, about 98%, or about 99%, sequence identity to the amino acid sequence set forth in SEQ ID NOs: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222. In other embodiments, the anti-SIRP α antibodies or antibody fragments thereof retain the binding and/or functional activity of an anti-SIRP α antibody or antibody fragment thereof that comprises the variable light chain sequence of SEQ ID NOs: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222. In still further embodiments, the anti-SIRP α antibodies or antibody fragments thereof comprise the variable light chain sequence of SEQ ID NOs: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222 and have one or more conservative amino acid substitutions, e.g., 1, 2, 3, 4, 5, 1-2, 1-3, 1-4 or 1-5 conservative amino acid substitutions in the light chain variable sequence. In yet further embodiments, the one or more conservative amino acid substitutions fall within one or more framework regions in SEQ ID NOs: 105, 106, 107, 108, 109, 127, 128, 129, 130, or 222 (based on the numbering system of Kabat).

[00194] In some embodiments, the anti-SIRP α antibody or antibody fragment thereof comprises a variable light chain sequence with at least about 95%, about 96%, about 97%, about 98%, or about 99% sequence identity to the anti-SIRP α light chain variable region sequence set forth in SEQ ID NOs: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222 comprises one or more conservative amino acid substitutions in a framework region (based on the numbering system of Kabat), and retains the binding and/or functional activity of an anti-SIRP α antibody or antibody fragment thereof that comprises a variable heavy chain sequence as set forth in SEQ ID NOs: 100, 101, 102, 103, 110, 111, 112, 113, 114, 115, 116, 117, 104, 118, 119, 120, 121, 122, 123, 124, or 221 and a variable light chain sequence as set forth in SEQ ID NOs: 105, 106, 107, 108, 109, 126, 127, 128, 129, 130, or 222.

[00195] In some embodiments, the present invention includes anti-SIRP α antibodies or antigen-binding fragments thereof having an amino acid substitution. These variants have at least one amino acid residue in the anti-SIRP α antibody or antigen-binding fragment thereof removed and a different residue inserted in its place. The sites of greatest interest for substitutional mutagenesis include the hypervariable regions, but FR alterations are also contemplated. Conservative substitutions are shown in Table 33 under the heading of "preferred substitutions". If such substitutions result in a change in biological activity, then more substantial changes, denominated "exemplary substitutions", or as further described below in reference to amino acid classes, may be introduced and the products screened.

Table 33: Exemplary amino acid substitutions

Original Residue	Exemplary Substitutions	Preferred Substitutions
Ala (A)	val; leu; ile	val
Arg (R)	lys; gln; asn	lys
Asn (N)	gln; his; asp, lys; arg	gln
Asp (D)	glu; asn	glu
Cys (C)	ser; ala	ser
Gln (Q)	asn; glu	asn
Glu (E)	asp; gln	asp
Gly (G)	ala	ala
His (H)	arg; asn; gln; lys;	arg
Ile (I)	leu; val; met; ala; phe; norleucine	leu
Leu (L)	ile; norleucine; val; met; ala; phe	ile
Lys (K)	arg; gln; asn	arg
Met (M)	leu; phe; ile	leu
Phe (F)	tyr; leu; val; ile; ala;	tyr
Pro (P)	ala	ala

Ser (S)	thr	thr
Thr (T)	ser	ser
Trp (W)	tyr; phe	tyr
Tyr (Y)	phe; trp; thr; ser	phe
Val (V)	leu; ile; met; phe ala; norleucine;	leu

In protein chemistry, it is generally accepted that the biological properties of the antibody can be accomplished by selecting substitutions that differ significantly in their effect on maintaining (a) the structure of the polypeptide backbone in the area of the substitution, for example, as a sheet or helical conformation, (b) the charge or hydrophobicity of the molecule at the target site, or (c) the bulk of the side chain. Naturally occurring residues are divided into groups based on common side-chain properties:

- (1) hydrophobic: norleucine, met, ala, val, leu, ile;
- (2) neutral hydrophilic: cys, ser, thr;
- (3) acidic: asp, glu;
- (4) basic: asn, gin, his, lys, arg;
- (5) residues that influence chain orientation: gly, pro; and
- (6) aromatic: trp, tyr, phe.

Non-conservative substitutions will entail exchanging a member of one of these classes for another class.

[00196] Any cysteine residue not involved in maintaining the proper conformation of the anti-SIRP α antibody or antigen-binding fragment thereof also may be substituted, generally with serine, to improve the oxidative stability of the molecule, prevent aberrant crosslinking, or provide for established points of conjugation to a cytotoxic or cytostatic compound. Conversely, cysteine bond(s) may be added to the antibody or antigen-binding fragment thereof to improve its stability (particularly where the antibody is an antibody fragment such as an Fv fragment).

[00197] Another type of amino acid variant of the antibody involves altering the original glycosylation pattern of the antibody. The term "altering" in this context means deleting one or more carbohydrate moieties found in the antibody, and/or adding one or more glycosylation sites that were not previously present in the antibody. For example, an antibody may comprise an

amino acid substitution at position 297 of the human IgG1 heavy chain to abrogate oligosaccharyltransferase enzyme complex-mediated glycosylation by replacing the asparagine 297 (e.g. N297A, N297G).

[00198] In some aspects, the present invention includes nucleic acid molecules that encode the amino acid sequence variants of the anti-SIRP α antibodies or antigen-binding fragments thereof described herein. Nucleic acid molecules encoding amino acid sequence variants of an anti-SIRP α antibody or antigen-binding fragment thereof are prepared by a variety of methods known in the art. These methods include, but are not limited to, isolation from a natural source (in the case of naturally occurring amino acid sequence variants) or preparation by oligonucleotide-mediated (or site-directed) mutagenesis, PCR mutagenesis, and cassette mutagenesis of an earlier prepared variant or a non-variant version of the anti-SIRP α antibody or antigen-binding fragment thereof. For example, nucleic acid molecules according to the invention also encompass nucleic acid molecules which hybridize under stringent conditions to nucleic acid molecules as disclosed herein, whereby the term "stringent conditions" within the scope of the invention can include, e.g., hybridization in a buffer comprising 50% formamide, 5 $\times$ SSC, and 1% SDS at 42 $^{\circ}$ C., or hybridization in a buffer comprising 5 $\times$ SSC and 1% SDS at 65 $^{\circ}$ C., both with a wash of 0.2 $\times$ SSC and 0.1% SDS at 65 $^{\circ}$ C. Exemplary stringent hybridization conditions can also include a hybridization in a buffer of 40% formamide, 1 M NaCl, and 1% SDS at 37 $^{\circ}$ C., and a wash in 1 $\times$ SSC at 45 $^{\circ}$ C.

[00199] In certain embodiments, the anti-SIRP α antibody is an antibody fragment. There are techniques that have been developed for the production of antibody fragments. Fragments can be derived via proteolytic digestion of intact antibodies (see, e.g., Morimoto et al., 1992, Journal of Biochemical and Biophysical Methods 24:107-117; and Brennan et al., 1985, Science 229:81). Alternatively, the fragments can be produced directly in recombinant host cells. For example, Fab'-SH fragments can be directly recovered from E. coli and chemically coupled to form F(ab')₂ fragments (see, e.g., Carter et al., 1992, Bio/Technology 10:163-167). By another approach, F(ab')₂ fragments can be isolated directly from recombinant host cell culture. Other techniques for the production of antibody fragments will be apparent to a skilled practitioner.

[00200] In one aspect, the anti-SIRP α antibodies and antigen-binding fragments thereof can include modifications, such as glycosylation, oxidation, or deamidation.

[00201] In certain embodiments, it may be desirable to use an anti-SIRP α antibody fragment, rather than an intact antibody. It may be desirable to modify the antibody fragment in

order to increase its serum half-life. This can be achieved, for example, by incorporation of a salvage receptor binding epitope into the antibody fragment. In one method, the appropriate region of the antibody fragment can be altered (e.g., mutated), or the epitope can be incorporated into a peptide tag that is then fused to the antibody fragment at either end or in the middle, for example, by DNA or peptide synthesis (see, e.g., WO 96/32478). For example, antibody fragments of the invention may also be fused to human serum albumin to increase the serum half-life, if the use of a full-length IgG scaffold is undesirable. Such fusion proteins of the antibody fragment with human serum albumin may be advantageous in situations in which two different antibody fragments need to be fused to increase avidity, or to generate a bispecific binding protein with extended serum half-life (see e.g. WO 05/077042 A2).

[00202] Removal of any carbohydrate moieties present on the antibody can be accomplished chemically or enzymatically. Chemical deglycosylation is described by Hakimuddin et al., 1987, Arch. Biochem. Biophys. 259:52 and by Edge et al., 1981, Anal. Biochem., 118:131. Enzymatic cleavage of carbohydrate moieties on antibodies can be achieved by the use of a variety of endo- and exo-glycosidases as described by Thotakura et al., 1987, Meth. Enzymol 138:350.

[00203] Another type of useful modification comprises linking the antibody to one of a variety of nonproteinaceous polymers, e.g., polyethylene glycol, polypropylene glycol, or polyoxyalkylenes, in the manner set forth in one or more of U.S. Pat. No. 4,640,835, U.S. Pat. No. 4,496,689, U.S. Pat. No. 4,301,144, U.S. Pat. No. 4,670,417, U.S. Pat. No. 4,791,192 and U.S. Pat. No. 4,179,337.

Biophysical Properties

[00204] In one aspect, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof having one or more favorable biophysical properties. In one aspect, a human anti-SIRP α antibody or antigen-binding fragment thereof of the present invention is present in at least 90% monomer form, or in at least 92% monomer form, or in at least 95% monomer form, or in at least 96% monomer form, or in at least 97% monomer form, or in at least 98% monomer form, or in at least 99% monomer form in a buffer. In a further aspect, a human anti-SIRP α antibody or antigen-binding fragment thereof of the present invention remains in at least 90% monomer form, or in at least 92% monomer form, or in at least 95% monomer form, or in at least 96% monomer form, or in at least 97% monomer form, or in at least 98% monomer form, or in at least 99% monomer form in a buffer for one week, one month, or for four months. Said percentage of monomer may be determined after one or more purification steps are

performed, e.g., Protein A purification optionally followed by cation exchange chromatography. Said percentage of monomer may be determined following a low pH treatment, e.g., pH 3.5 treatment. Said percentage of monomer may be determined following one week, one month, or four months in a buffer at room temperature, e.g., 25° C., or an elevated temperature, e.g., 40° C.

[00205] In another aspect, an anti-SIRP α antibody or antigen-binding fragment thereof of the invention is stable at high concentrations.

[00206] In another aspect, an anti-SIRP α antibody or antigen-binding fragment thereof of the invention has low viscosity.

Epitope binding

[00207] In another aspect, the invention relates to an antibody or antigen-binding fragment thereof that recognizes a specific linear and/or conformational "SIRP α antigen epitope" and "SIRP α epitope".

[00208] As used herein, the terms "SIRP α antigen epitope" and "SIRP α epitope" refer to a molecule (e.g., a peptide) or a fragment of a molecule capable of binding to an anti-SIRP α antibody or antigen-binding fragment thereof. These terms further include, for example, a SIRP α antigenic determinant recognized by any of the antibodies or antibody fragments of the present invention or key points of contact between the molecule and antibody.

[00209] SIRP α antigen epitopes can be included in proteins, protein fragments, peptides or the like. The epitopes are most commonly proteins, short oligopeptides, oligopeptide mimics (e.g., organic compounds that mimic antibody binding properties of the SIRP α antigen), or combinations thereof.

[00210] In one aspect of the invention, the antibody or antigen-binding fragment thereof recognizes a specific linear or conformational "SIRP α epitope" on a SIRP α protein comprising the amino acid sequence set forth in any one of SEQ ID NOs: 240-252. In one aspect of the invention, the antibody or antigen-binding fragment thereof recognizes a specific linear or conformational "SIRP α epitope" on a SIRP α protein comprising the amino acid sequence set forth in any one of SEQ ID NOs: 253-260 and 264, in particular any one or more of SEQ ID NOS: 256 and 257. In one aspect of the invention the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising the amino acids: LEU 60, ILE 61, VAL 63, GLY 64, PRO 65, GLN 82, LYS 83, GLU 84, THR 97, LYS 98, ARG 99, GLU 100, LYS 126, GLY 127, SER 128, PRO 129 and ASP

130 as set forth in SEQ ID NO: 266. In one aspect of the invention the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising the amino acids LEU 60, ILE 61, VAL 63, GLY 64, PRO 65, GLN 82, LYS 83, GLU 84, THR 97, LYS 98, ARG 99, ASN 100, LYS 126, GLY 127, SER 128, PRO 129 and ASP 130 as set forth in SEQ ID NO: 265. In another aspect of the invention, the antibody or antigen-binding fragment thereof binds to the SIRP α epitope comprising the amino acid ASP130. In another aspect, the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising any one or more of SEQ ID NO: 253, SEQ ID NO: 254, SEQ ID NO: 255, SEQ ID NO: 264, and SEQ ID NO: 256, in particular SEQ ID NO: 256.

[00211] In one aspect of the invention the antibody or antigen-binding fragment thereof binds to the SIRP α epitope comprising the amino acids ARG 70, GLY 71, ALA 72, GLY 73, PRO 74, ALA 75, ARG 76, GLU 77, ALA 114, ALA 116, GLY 117, THR 118, TYR 120, THR 131, GLU 132, PHE 133, SER 135 and GLU 140 as set forth in SEQ ID NO: 266. In one aspect of the invention the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising the amino acids ARG 70, GLY 71, ALA 72, GLY 73, PRO 74, GLY 75, ARG 76, GLU 77, ALA 114, ALA 116, GLY 117, THR 118, TYR 120, VAL 132, GLU 133, PHE 134, SER 136 and GLU 141 as set forth in SEQ ID NO: 265. In another aspect of the invention, the antibody or antigen-binding fragment thereof binds to the SIRP α epitope comprising the amino acid ALA72. In another aspect, the antibody or antigen-binding fragment thereof binds to a SIRP α epitope comprising any one or more of SEQ ID NO: 257, SEQ ID NO: 258, SEQ ID NO: 259 and SEQ ID NO: 260, in particular SEQ ID NO: 257. In another aspect of the invention, the antibody or antigen-binding fragment thereof blocks binding of any of Antibody A-E to SIRP α .

[00212] In a further embodiment, the epitope is present on a SIRP α protein set forth in Table 34 or 35 below.

Table 34: Sequences of SIRP proteins

Protein	Sequence (Extracellular Domain)	SEQ ID NO:
Human SIRP α V1 (NP_542970.1)	EEELQVIQPDKSVLVAAGETATLRCTATSLIPVGPIQWFR GAGPGRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRIG NITPADAGTYCYVKFRKGSPDDVEFKSGAGTELSVRAKPS APVVS GPAARATPQHTVSFTCESHGFSRPDITLKWFKNG NELSDFQTNVDPVGESVSYSIHSTAKVVLTREDVHSQVIC EVAHVTLQGDPLRG TANLSETIRVPPTLEVTQQPVRAEN QVNVTCQVRKFYPQRLQLTWLENGNVSRTETASTVTEN KDGTYNWMMSWLLVNVSAHRDDVKLTCQVEHDGQPAV SKSHDLKVS AHPKEQGSNTAAENTGSNERNIYGGSG	240
Human SIRP α V2 (CAA71403.1)	EEELQVIQPDKSVSVAAGESAILHCTVTSIPVGPIQWFRG AGPARELIYNQKEGHFPRVTTVSESTKRENMDFSISINIT PADAGTYCYVKFRKGSPDTEFKSGAGTELSVRAKPSAPVV SGPAARATPQHTVSFTCESHGFSRPDITLKWFKNGNELS DFQTNVDPVGESVSYSIHSTAKVVLTREDVHSQVICEVAH VTLQGDPLRG TANLSETIRVPPTLEVTQQPVRAENQVNV TCQVRKFYPQRLQLTWLENGNVSRTETASTVTENKDGTY NWMMSWLLVNVSAHRDDVKLTCQVEHDGQPAVSKSHDL KVS AHPKEQGSNTAAENTGSNERNIYGGSG	241
Human SIRP β 1 (NP_006056.2).	EDELQVIQPEKSVSVAAGESATLRCAMTSLIPVGPIMWFR GAGAGRELIYNQKEGHFPRVTTVSELTKRNNLDFSISINI TPADAGTYCYVKFRKGSPDDVEFKSGAGTELSVRAKPSAP VVS GPAVRATPEHTVSFTCESHGFSRPDITLKWFKNGNEL SDFQTNVDPAGDSVSYSIHSTARVVLTRGDVHSQVICEIA HITLQGDPLRG TANLSEAIRVPPTLEVTQQPMRAENQAN VTCQVSNFYPRGLQLTWLENGNVSRTETASTLIENKDGTY YNWMMSWLLVNTCAHRDDVVLTCQVEHDGQQA VSKSY ALEISAHQKEHGS DITHEAALAPTAPLGGSG	242
Human SIRP β L/SIRP β 1v3 (NP_001129316.1)	EEELQVIQPDKSVSVAAGESATLHCTVTSIPVGPIQWFRG AGPGRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRISNI TPADAGTYCYVKFRKGSPDHVEFKSGAGTELSVRAKPSAP VVS GPAARATPQHTVSFTCESHGFSRPDITLKWFKNGNE LSDFQTNVDPAGDSVSYSIHSTAKVVLTREDVHSQVICEV AHVTLQGDPLRG TANLSETIRVPPTLEVTQQPVRAENQV NVTCQVRKFYPQRLQLTWLENGNVSRTETASTLTENKDG TYNWMMSWLLVNVSAHRDDVKLTCQVEHDGQPAVSKSH DLKVS AHPKEQGSNTAPGPALASAAPLGGSG	243

human SIRP γ (NP_061026.2)	EEELQMIQPEKLLLVTVGKTATLHCTVTSLPVG PVLWFR GVGPGRELIYNQKEGHFPRVTVSDLTRNNMDFSIRISSI TPADVGTYYCVKFRKGSPENVEFKSGPGTEMALGAKPSA PVVLGPAARTTPEHTVSFTCESHGFSRPDITLKWFKNGNE LSDFQTNVDPTGQSVAYSIRSTARVVLDPWDVRSQVICE VAHVTLQGDPLRG TANLSEAIRVPPTLEV TQQPMRVGN QVNVTCQVRKFYPQSLQLTWS ENGNVCQRETASTLTEN KDGTYNWT SWFLVNISDQRDDVLT CQVKHDGQLAVS KRLALEVT VH QKDQSSD ATP	244
Human CD47 (NP_942088)	WQPPLL FNKTSVEFTFGNDTVVIPCFTNMEA QNTTEV YVKWKF KGRDIYTFD GQANKSTVPTDFSSAKIEVS QLLKG DASLKMDKSDAVSHTGNYTCEVTELTREGETIIE LKYRVVS	245
Mouse SIRP α (NP_031573)	KELKV TQPEKSVSVAAGDSTVLNCTLSLLPVGPIKWYRG VGQSRLIYSFTGEHFPRVTNVSDATKRNNMDFSIRISNV TPEDAGTYYCVKFQKGPSEPDEIQSGGGTEVYVLAKPSP PEVSGPADRGIPDQKVNFTCKSHGFSRNITLKWFKDGQ ELHHLETTVNPSGKNVSYNISSTVRVVLNSMDVH SKVICE VAHITLDRSPLRGIANLSNFIRVSPTVKVTQQSPTSMNQV NLTCRAERFYPEDLQLIWL ENGNVSRNDTPKNLTKN TDG TYNYTSLFLVNSSAHREDVFT CQVKHDQQPAITRNHTV LGLAHSSDQGSMTFPGNNATHNWN	246
CySIRP α NP_001271679	EEELQVIQPEKSVSVAAGDSATLNCTVSSLIPVGPIQWFR GAGPGRELIYNLKEGHFPRVTAVSDPTKRNNMDFSIRISN ITPADAGTYYCVKFRKGSPDVELKSGAGTELSVR AKPSAP VVSGPAVRATAEHTVSFTCESHGFSRPDITLKWFKNGNEL SDVQTNVDPAGKSVSY SIRSTARVLLTRRDVHSQVICEVA HVTLQGDPLRG TANLSEAIRVPPFLEV TQQSMRADNQV NVTCQVTKFYPQRLQLTWLENGNVSRTEMASALPENKD GTYNWT SWLLVNVS AHRDDVKLT CQVEHDGQPAVNKS FSVKVSAHPKEQGSNTAAENTGTNERNIYGGSG	247
CySIRP α EGM_02252	EEELQVIQPEKSVSVAAGESATLNCTATSLIPVGPIQWFR GVGPGRELIYHQKEGHFPRVTPVSDPTKRNNMDFSIRISN ITPADAGTYYCVKFRKGSPDVELKSGAGTELSVR AKPSAP VVSGPAVRATAEHTVSFTCESHGFSRPDITLKWFKNGNEL SDFQTNVDPAGKSVSY SIRSTARVVLTRRDVHSQVICEVA HVTLQGDPLRG TANLSEAIRVPPFLEF TQQSMRADNQV NVTCQVMKFYPQRLQLTWLENGNVSRTEMASALPENK DGTYNWT SWLLVNVS AHRDDVKLT CQVEHDGQPAVNK SFSVKVSAHPKEQGSNTAAENTGTNERNIYGGSG	248

CySIRP α XP_015313155	EEELQVIQPEKSVSVAAGDSATLNCTVSSLIPVGPIQWFR GAGPGRELIYNLKEGHFPRVTPVSDPTKRNNMDFSIRISN ITPADAGTYICVKFRKGSPDVELKSGAGTELSVRAPKSAP VVSGPAVRATAEHTVSFTCESHGFSRPDITLKWFKNGNEL SDFQTNVDPAGKSVSYSIRSTARVVLTRRDVHSQVICEVA HVTLQGDPLRGATANLSEAIRVPPFLEVTTQSMRADNQV NVTCQVTKFYPQRLQLTWLENGNVSRTEMASALPENKD GTYNWTSWLLVNVSAHRDDVKLTCQVEHDGQPAVNKS FSVKVSAHPKEQGSNTAAENTGTNERNIYGGSG	249
Cyno SIRP β 1v3 XP_005568593	EEELQVIQPEKSVSVTAGESATLNCTVSLIPVGPIQWFRG AGPGRELIYNLKEGHFPRVTTVLDPDKRNNMDFSIRISNIT PADAGTYICVKFRKGSPDVELKSGAGTELSVRAPKSAPVV SGPTARATPEHTVSFTCESHGFSRPDITLKWFKNGNELSD FQTNVDPAGKSVSYSIRSTARVVLTRRDVHSQVICEVAHV TLQGDPLRGATANLSEAIRVPPTEVFQRPMAENQVNV CQVRKFYPQRLLLTWLENGNVSTETASTLTENKDGTYN WRSWLLVNTCAHRDGVVLTQVEHDGQPAVSKSHALE VSAHQKEQCSDTTSGPVLAPTAPLGGSG	250
Cyno SIRP β 1 XP_005568598	EEELQVIQPEKSVSVAAGESATLNCTVSLIPVGPIQWFRG VGPGRELIFNLKEGHFPRVTPVSDPTKRNNMDFSILISSIT PADAGTYICVKFRKGSPDVELKSGAGTELSVRAPKSAPVV SGPAVRATAEHTVSFTCESHGFSRPDITLKWFKNGNELSD FQTSVDPAGKSVSYSIRSTARVVLTRRDVHSQVICEVAHV TLQGDPLRGATANLSEAIRVPPFLEVTTQSMRAENQANIT CQVSNFYQRLLLTWLENGNVSTETASTLTENKDGTYN WTSWLLVNICHRDDVLTQVKHGDGQPAVSKSHTLEIS AHQKEQSDSVTHGLALAPTAPLGGSG	251
Cyno SIRP γ XP_005568591	EEELQMIQPEKLLLVAVGESATLNCTVTSLLPVGPIVWFR GVGPGRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRISSI TPADAGTYICVKFRKGSPENVEFKSGPGTEMALRAKPSA PVVSGPAARATPEHTVSFTCKSHGFSRPDITLKWFKNGN ELSDFTNVDPAGQSVSYSIRSTARVVLAPWDVRSQVTC EVAHVTLQGDPLRGATANLSEAIRVPPTEVTQQPMRAG NQVNITYQVRNFYPQNLQLTWLENGNVCRTEASTLTEN KDGTYNWTWLLVNTSDQRDDVLTQVKHGDGQLAVN KSLVLEVSAHQKQSSDATHGGSG	252

Table 35: Sequences of SIRP α epitopes

Protein	Sequence	SEQ ID NO:
Human SIRP α V1 or V2	LIPVGP	253
Human SIRP α V1 or V2	QKE	254

Human SIRP α V1	TKRN	255
Human SIRP α V2	TKRE	264
Human SIRP α V1 or V2	KGSPD	256
Human SIRP α V1 or V2	RGAGPGRE	257
Human SIRP α V1 or V2	AGTYT	258
Human SIRP α V1	VEFKSGAGTE	259
Human SIRP α V2	TEFKSGAGTE	260

[00213] The present invention also provides an anti-SIRP α antibody or antigen-binding fragment thereof that competes for binding to SIRP α with an anti-SIRP α antibody according to the present invention. In one embodiment, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof that competes for binding to SIRP α with any one of Antibody A-Antibody E. Competition assays may be conducted for example as described in PLoS One. 2014; 9(3): e92451 using a biosensor, or PLoS One 2020 Mar 5;15(3):e0229206, or by a method disclosed herein.

Therapeutic Uses

[00214] In one embodiment, the anti-SIRP α antibodies of the invention or antigen-binding fragments thereof are useful for treating and/or preventing SIRP α pathway disorders.

[00215] In another embodiment, the anti-SIRP α antibodies of the invention or antigen-binding fragments thereof are useful as a medicament.

[00216] Accordingly, in one embodiment, the invention provides a method of modulating the interaction between SIRP α and CD47 in a subject comprising administering to said subject a composition comprising an anti-SIRP α antibody or antigen-binding fragment thereof according to the invention in an amount sufficient to block CD47-mediated SIRP α signaling in said subject. In one embodiment, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof according to the present invention for use in modulating the interaction between SIRP α and CD47 in a subject. In one embodiment, the invention provides the use of an anti-SIRP α antibody or antigen-binding fragment thereof

according to the present invention in the manufacture of a medicament for modulating the interaction between SIRP α and CD47 in a subject.

[00217] In one embodiment, the invention provides a method of enhancing myeloid cell phagocytosis in a subject comprising administering to said subject a composition comprising an anti-SIRP α antibody or antigen-binding fragment thereof according to the invention in an amount sufficient to enhance an immune response in said subject. In one embodiment, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof according to the invention for use in enhancing myeloid cell activity in a subject. In one embodiment, the present invention provides the use of an anti-SIRP α antibody or antigen-binding fragment thereof according to the present invention in the manufacture of a medicament for enhancing myeloid cell phagocytosis in a subject.

[00218] In one embodiment, a SIRP α pathway disease or disorder in a subject comprising administering to said subject a composition comprising an anti-SIRP α antibody or antigen-binding fragment thereof according to the present invention. In one embodiment, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof according to the present invention for use in treating or preventing cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis in a subject. In one embodiment, the present invention provides the use of an anti-SIRP α antibody or antigen-binding fragment thereof according to the present invention in the manufacture of a medicament for treating or preventing cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis in a subject.

[00219] Accordingly, in one embodiment, the invention provides a method of treating or preventing one of the above diseases or disorders in a subject comprising administering to said subject a composition comprising an anti-SIRP α antibody or antigen-binding fragment thereof according to the invention. In one embodiment, the present invention provides an anti-SIRP α antibody or antigen-binding fragment thereof according to the invention for use in treating or preventing one of the above diseases or disorders in a subject. In one embodiment, the present invention provides the use of an anti-SIRP α antibody or antigen-binding fragment thereof according to the present invention in the manufacture of a medicament for treating and/or preventing one of the above diseases or disorders in a subject.

Non-Therapeutic Uses

[00220] The antibodies described herein are useful as affinity purification agents. In this process, the antibodies are immobilized on a solid phase such as a Protein A resin, using methods well known in the art. The immobilized antibody is contacted with a sample containing the SIRP α protein (or fragment thereof) to be purified, and thereafter the support is washed with a suitable solvent that will remove substantially all the material in the sample except the SIRP α protein, which is bound to the immobilized antibody. Finally, the support is washed with another suitable solvent that will release the SIRP α protein from the antibody.

[00221] The SIRP α antibodies and fragments thereof of the invention as disclosed herein are also useful in diagnostic assays to detect and/or quantify SIRP α protein, for example, detecting SIRP α expression in specific cells, tissues, or serum.

[00222] It will be advantageous in some embodiments, for example, for diagnostic purposes to label the antibody with a detectable moiety. Numerous detectable labels are available, including radioisotopes, fluorescent labels, enzyme substrate labels, quantum dots and the like. The label may be indirectly conjugated with the antibody using various known techniques. For example, the antibody can be conjugated with biotin and any of the three broad categories of labels mentioned above can be conjugated with avidin, or vice versa. Biotin binds selectively to avidin and thus, the label can be conjugated with the antibody in this indirect manner. Alternatively, to achieve indirect conjugation of the label with the antibody, the antibody can be conjugated with a small hapten (such as digoxin) and one of the different types of labels mentioned above is conjugated with an anti-hapten antibody (e.g., anti-digoxin antibody). Thus, indirect conjugation of the label with the antibody can be achieved.

Diagnostic Kits

[00223] An anti-SIRP α antibody or fragment thereof can be used in a diagnostic kit, i.e., a packaged combination of reagents in predetermined amounts with instructions for performing the diagnostic assay. Where the antibody is labeled with an enzyme, the kit may include substrates and cofactors required by the enzyme such as a substrate precursor that provides the detectable chromophore or fluorophore. In addition, other additives may be included such as stabilizers, buffers (for example a block buffer or lysis buffer), and the like. The relative amounts of the various reagents may be varied widely to provide for concentrations in solution of the reagents that substantially optimize the sensitivity of the assay. The reagents may be provided

as dry powders, usually lyophilized, including excipients that on dissolution will provide a reagent solution having the appropriate concentration.

Compositions, Combinations and Administration Thereof

[00224] A composition comprising an anti-SIRP α antibody or an antigen-binding fragment thereof according to the invention can be administered to a subject having or at risk of the SIRP α pathway diseases and/or disorders described herein. The invention further provides for the use of an anti-SIRP α antibody or an antigen-binding fragment thereof in the manufacture of a medicament for prevention or treatment of a SIRP α pathway disease or disorder. The term "subject" as used herein means any mammalian patient to which an anti-SIRP α antibody or an antigen-binding fragment thereof can be administered, including, e.g., humans and certain non-human mammals, such as primates, and dogs. Subjects specifically intended for treatment using the methods described herein include humans.

[00225] An anti-SIRP α antibody or an antigen-binding fragment thereof may be administered on their own or in combination with one or more additional therapeutic agents, such as state-of-the-art or standard-of-care compounds, such as e.g. cytostatic or cytotoxic substances, cell proliferation inhibitors, anti-angiogenic substances, steroids, immune modulators / checkpoint inhibitors, and the like.

[00226] In one aspect, the present invention also provides pharmaceutical compositions administered as pharmaceutical compositions comprising a therapeutically effective amount of the anti-SIRP α antibody or an antigen-binding fragment thereof and one or more pharmaceutically compatible ingredients, and optionally one or more additional therapeutic agents.

[00227] A further aspect of the invention provides a binding molecule of the invention for use in the therapy of cancer (e.g. an individual suffering from cancer or being at risk of developing cancer) wherein said therapy comprises one or more pharmacologically active substances.

[00228] A further aspect of the invention provides the use of one or more active ingredients in the manufacture of a medicament for the therapy of cancer and/or tumors (e.g. an individual suffering from cancer or being at risk of developing cancer) wherein said medicament comprises the binding molecule of the invention.

[00229] Cytostatic and/or cytotoxic active substances which may be administered in combination with an anti-SIRP α antibody or antigen-binding fragment thereof of the invention include, without being restricted thereto, hormones, hormone analogues and anti-hormones, aromatase inhibitors, LHRH agonists and antagonists, inhibitors of growth factors (e.g., platelet derived growth factor (PDGF), fibroblast growth factor (FGF), vascular endothelial growth factor (VEGF), epidermal growth factor (EGF), insulin-like growth factors (IGF), human epidermal growth factor (HER, e.g. HER2, HER3, HER4) and hepatocyte growth factor (HGF)), including, for example anti-growth factor antibodies or anti-growth factor receptor antibodies and tyrosine kinase inhibitors, such as, for example, cetuximab, gefitinib, afatinib, nintedanib, imatinib, lapatinib, bosutinib and trastuzumab; antimetabolites (e.g., antifolates such as methotrexate, raltitrexed, pyrimidine analogues such as 5-fluorouracil (5-FU), gemcitabine, irinotecan, doxorubicin, TAS-102, capecitabine and gemcitabine, purine and adenosine analogues such as mercaptopurine, thioguanine, cladribine and pentostatin, cytarabine (ara C), fludarabine); antitumor antibiotics (e.g., anthracyclins); platinum derivatives (e.g., cisplatin, oxaliplatin, carboplatin); alkylation agents (e.g., estramustin, meclorethamine, melphalan, chlorambucil, busulphan, dacarbazine, cyclophosphamide, ifosfamide, temozolomide, nitrosoureas such as for example carmustine and lomustine, thiotepa); antimetotic agents (e.g., Vinca alkaloids such as for example vinblastine, vindesine, vinorelbine and vincristine; and taxanes such as paclitaxel, docetaxel); angiogenesis inhibitors, including bevacizumab, ramucirumab and aflibercept, tubulin inhibitors; DNA synthesis inhibitors, PARP inhibitors, topoisomerase inhibitors (e.g., epipodophyllotoxins such as for example etoposide and etopophos, teniposide, amsacrine, topotecan, irinotecan, mitoxantrone), serine/threonine kinase inhibitors (e.g., PDK1 inhibitors, Raf inhibitors, A-Raf inhibitors, B-Raf inhibitors, C-Raf inhibitors, mTOR inhibitors, mTORC1/2 inhibitors, PI3K inhibitors, PI3K α inhibitors, dual mTOR/PI3K inhibitors, STK33 inhibitors, AKT inhibitors, PLK1 inhibitors (such as volasertib), inhibitors of CDKs, including CDK9 inhibitors, Aurora kinase inhibitors), tyrosine kinase inhibitors (e.g., PTK2/FAK inhibitors), protein interaction inhibitors, MEK inhibitors, ERK inhibitors, FLT3 inhibitors, BRD4 inhibitors, IGF-1R inhibitors, Bcl-xL inhibitors, Bcl-2 inhibitors, Bcl-2/Bcl-xL inhibitors, ErbB receptor inhibitors, BCR-ABL inhibitors, ABL inhibitors, Src inhibitors, rapamycin analogs (e.g., everolimus, temsirolimus, ridaforolimus, sirolimus), androgen synthesis inhibitors, androgen receptor inhibitors, DNMT inhibitors, HDAC inhibitors, ANG1/2 inhibitors, CYP17 inhibitors, radiopharmaceuticals, immunotherapeutic agents such as immune checkpoint inhibitors (e.g., CTLA4, PD1, PD-L1, LAG3, and TIM3 binding molecules / immunoglobulins, such as ipilimumab, nivolumab, pembrolizumab) and various chemotherapeutic agents such as amifostin, anagrelide, clodronate, filgrastin, interferon, interferon

alpha, leucovorin, rituximab, procarbazine, levamisole, mesna, mitotane, pamidronate and porfimer; proteasome inhibitors (such as Bortezomib); Smac and BH3 mimetics; agents restoring p53 functionality including mdm2-p53 antagonist; inhibitors of the Wnt/beta-catenin signaling pathway; and/or cyclin-dependent kinase 9 inhibitors.

[00230] In one aspect of the invention, the anti-SIRP α antibody or antigen-binding fragment thereof is optionally administered in combination with an additional therapeutic agent. The additional therapeutic agent may be a chemotherapeutic agent, an anti-PD-1 or PD-L1 antibody, an anti-CTLA4 antibody, a T-cell engager, a CD137-agonist-anti-FAP bispecific antibody, a tumor-targeting antibody, a VEGF-ANG2 bispecific antibody, a STING agonist, a MDM2 antagonist, or radiation therapy.

[00231] In one aspect, the anti-SIRP α antibody or an antigen-binding fragment thereof is administered in combination with an anti-PD-1 antibody, for example, nivolumab, pembrolizumab, pidilizumab, ezabenlimab, or atezolizumab. In a further aspect, the anti-SIRP α antibody or an antigen-binding fragment thereof is administered in combination with an anti-PD-L1 antibody including, for example, avelumab or durvalumab.

[00232] In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof is administered in combination with a tumor targeting antibody targeting HER2 (e.g., trastuzumab), EGFR (e.g., cetuximab, panitumumab), CD20 (e.g., rituximab, ofatumumab), or CD52 (e.g., alemtuzumab).

[00233] In one aspect, the anti-SIRP α antibody or antigen-binding fragment thereof is administered in combination with two therapeutic agents. In a further aspect, the anti-SIRP α antibody or antigen-binding fragment thereof is administered in combination with an anti-PD-1 antibody (e.g., nivolumab, pembrolizumab, pidilizumab, ezabenlimab, or atezolizumab), or an anti-PD-L1 antibody (e.g., avelumab or durvalumab) and tumor targeting antibody targeting HER2 (e.g., trastuzumab), EGFR (e.g., cetuximab, panitumumab), CD20 (e.g., rituximab, ofatumumab), or CD52 (e.g., alemtuzumab).

[00234] Various delivery systems are known and can be used to administer the anti-SIRP α antibody or an antigen-binding fragment thereof. Methods of introduction include but are not limited to intravitreal, eye drops, intradermal, intramuscular, intraperitoneal, intravenous, subcutaneous, intranasal, epidural, and oral routes. The anti-SIRP α antibody or an antigen-binding fragment thereof can be administered, for example by infusion, bolus or injection, and can be administered together with other biologically active agents. Administration can be systemic or

local. Formulations for such injections may be prepared in, for example, prefilled syringes. As such in an aspect of the invention, pre-filled syringes are provided that include an anti-SIRP α antibody or an antigen-binding fragment thereof.

[00235] To be used in therapy, the anti-SIRP α antibody of the invention is formulated into pharmaceutical compositions appropriate to facilitate administration to animals or humans. Typical formulations of the binding molecule or antibody molecule described herein can be prepared by mixing the binding molecule or antibody molecule with physiologically acceptable carriers, excipients or stabilizers, in the form of lyophilized or otherwise dried formulations or aqueous solutions or aqueous or non-aqueous suspensions. Carriers, excipients, modifiers or stabilizers are nontoxic at the dosages and concentrations employed.

[00236] In typical embodiments, the pharmaceutical composition is formulated in accordance with routine procedures as a pharmaceutical composition adapted for intravenous or subcutaneous administration to a subject. Typically, compositions for administration by injection are solutions in sterile isotonic aqueous buffer. Where necessary, the pharmaceutical composition can also include a solubilizing agent and a local anesthetic such as lignocaine to ease pain at the site of the injection. Generally, the ingredients are supplied either separately or mixed together in unit dosage form, for example, as a dry lyophilized powder or water free concentrate in a hermetically sealed container such as an ampoule or sachet indicating the quantity of active agent. Where the pharmaceutical is to be administered by infusion, it can be dispensed with an infusion bottle containing sterile pharmaceutical grade water or saline. Where the pharmaceutical is administered by injection, an ampoule of sterile water for injection or saline can be provided so that the ingredients can be mixed prior to administration.

[00237] Further, the pharmaceutical composition can be provided as a pharmaceutical kit comprising (a) a container containing an anti-SIRP α antibody or an antigen-binding fragment thereof in lyophilized form and (b) a second container containing a pharmaceutically acceptable diluent (e.g., sterile water) for injection. The pharmaceutically acceptable diluent can be used for reconstitution or dilution of the lyophilized anti-SIRP α antibody or antigen-binding fragment thereof. Optionally associated with such container(s) can be a notice in the form prescribed by a governmental agency regulating the manufacture, use or sale of pharmaceuticals or biological products, which notice reflects approval by the agency of manufacture, use or sale for human administration.

[00238] In some embodiments, antibodies of the invention can be formulated to doses, which include for example a dose of 1 mg/kg to 200 mg/kg. including, for example, 1mg/kg to 25 mg/kg, 25 mg/kg to 50 mg/kg, 50 mg/kg to 75 mg/kg, 75 mg/kg to 100 mg/kg, 100 mg/kg to 125 mg/kg, 125 mg/kg to 150 mg/kg, 150 mg/kg to 175 mg/kg, or 175 mg/kg to 200 mg/kg. With respect to therapeutic regimens for combinatorial administration, in a specific embodiment, an anti-SIRP α antibody or antigen-binding fragment thereof is administered concurrently with a second and/or third therapeutic agent. In another specific embodiment, the second and/or third therapeutic agent is administered prior or subsequent to administration of the anti-SIRP α antibody or antigen-binding fragment thereof.

Polynucleotides, Vectors, and Host Cells

[00239] The present invention relates to isolated polynucleotides that comprise a sequence encoding an anti-SIRP α antibody or antigen-binding fragment thereof, vectors, and host cells comprising the polynucleotides, and recombinant techniques for production of the antibody. The isolated polynucleotides can encode any desired form of the anti-SIRP α antibody including, for example, full length monoclonal antibodies, Fab, Fab', F(ab')₂, and Fv fragments, diabodies, linear antibodies, single-chain antibody molecules, and multispecific antibodies formed from antibody fragments.

[00240] The polynucleotide(s) that comprise a sequence encoding an anti-SIRP α antibody or a fragment or chain thereof can be fused to one or more regulatory or control sequence, as known in the art, and can be contained in suitable expression vectors or host cell as known in the art. Each of the polynucleotide molecules encoding the heavy or light chain variable domains can be independently fused to a polynucleotide sequence encoding a constant domain, such as a human constant domain, enabling the production of intact antibodies. Alternatively, polynucleotides, or portions thereof, can be fused together, providing a template for production of a single chain antibody.

[00241] For recombinant production, a polynucleotide encoding the antibody is inserted into a replicable vector for cloning (amplification of the DNA) or for expression. Many suitable vectors for expressing the recombinant antibody are available. The vector components generally include, but are not limited to, one or more of the following: a signal sequence, an origin of replication, one or more marker genes, an enhancer element, a promoter, and a transcription termination sequence.

[00242] The anti-SIRP α antibodies can also be produced as fusion polypeptides, in which the antibody is fused with a heterologous polypeptide, such as a signal sequence or other polypeptide having a specific cleavage site at the amino terminus of the mature protein or polypeptide. The heterologous signal sequence selected is typically one that is recognized and processed (e.g., cleaved by a signal peptidase) by the host cell. For prokaryotic host cells that do not recognize and process the anti-SIRP α antibody signal sequence, the signal sequence can be substituted by a prokaryotic signal sequence. The signal sequence can be, for example, alkaline phosphatase, penicillinase, lipoprotein, heat-stable enterotoxin II leaders, and the like. For yeast secretion, the native signal sequence can be substituted, for example, with a leader sequence obtained from yeast invertase alpha-factor (including *Saccharomyces* and *Kluyveromyces* α -factor leaders), acid phosphatase, *C. albicans* glucoamylase, or the signal described in WO 90/13646. In mammalian cells, mammalian signal sequences as well as viral secretory leaders, for example, the herpes simplex gD signal, can be used. The DNA for such precursor region is ligated in reading frame to DNA encoding the anti-SIRP α antibody.

[00243] Anti-SIRP α antibody transcription from vectors in mammalian host cells is controlled, for example, by promoters obtained from the genomes of viruses such as polyoma virus, fowlpox virus, adenovirus (such as Adenovirus 2), bovine papilloma virus, avian sarcoma virus, cytomegalovirus, a retrovirus, hepatitis-B virus and Simian Virus 40 (SV40), from heterologous mammalian promoters, e.g., the actin promoter or an immunoglobulin promoter, or from heat-shock promoters, provided such promoters are compatible with the host cell systems.

[00244] Suitable host cells for cloning or expressing the DNA in the vectors herein are the prokaryote, yeast, or higher eukaryote cells described above. Suitable prokaryotes for this purpose include eubacteria, such as Gram-negative or Gram-positive organisms, for example, Enterobacteriaceae such as *Escherichia*, e.g., *E. coli*, *Enterobacter*, *Erwinia*, *Klebsiella*, *Proteus*, *Salmonella*, e.g., *Salmonella typhimurium*, *Serratia*, e.g., *Serratia marcescans*, and *Shigella*, as well as Bacilli such as *B. subtilis* and *B. licheniformis* (e.g., *B. licheniformis* 41 P disclosed in DD 266,710 published Apr. 12, 1989), *Pseudomonas* such as *P. aeruginosa*, and *Streptomyces*. One preferred *E. coli* cloning host is *E. coli* 294 (ATCC 31,446), although other strains such as *E. coli* B, *E. coli* X1776 (ATCC 31,537), and *E. coli* W3110 (ATCC 27,325) are suitable. These examples are illustrative rather than limiting.

[00245] In addition to prokaryotes, eukaryotic microbes such as filamentous fungi or yeast are suitable cloning or expression hosts for anti-SIRP α antibody-encoding vectors. *Saccharomyces cerevisiae*, or common baker's yeast, is the most commonly used among lower

eukaryotic host microorganisms. However, a number of other genera, species, and strains are commonly available and useful herein, such as *Schizosaccharomyces pombe*; *Kluyveromyces* hosts such as, e.g., *K. lactis*, *K. fragilis* (ATCC 12,424), *K. bulgaricus* (ATCC 16,045), *K. wickerhamii* (ATCC 24,178), *K. waltii* (ATCC 56,500), *K. drosophilae* (ATCC 36,906), *K. thermotolerans*, and *K. marxianus*; *Yarrowia* (EP 402,226); *Pichia pastoris* (EP 183,070); *Candida*; *Trichoderma reesei* (EP 244,234); *Neurospora crassa*; *Schwanniomyces* such as *Schwanniomyces occidentalis*; and filamentous fungi such as, e.g., *Neurospora*, *Penicillium*, *Tolypocladium*, and *Aspergillus* hosts such as *A. nidulans* and *A. niger*.

[00246] Suitable host cells for the expression of glycosylated anti-SIRP α antibody are derived from multicellular organisms. Examples of invertebrate cells include plant and insect cells, including, e.g., numerous baculoviral strains and variants and corresponding permissive insect host cells from hosts such as *Spodoptera frugiperda* (caterpillar), *Aedes aegypti* (mosquito), *Aedes albopictus* (mosquito), *Drosophila melanogaster* (fruitfly), and *Bombyx mori* (silkworm). A variety of viral strains for transfection are publicly available, e.g., the L-1 variant of *Autographa californica* NPV and the Bm-5 strain of *Bombyx mori* NPV, and such viruses may be used, particularly for transfection of *Spodoptera frugiperda* cells.

[00247] Plant cell cultures of cotton, corn, potato, soybean, petunia, tomato, and tobacco can also be utilized as hosts.

[00248] The anti-SIRP α antibodies or antigen-binding fragments thereof can also be incorporated in viral vectors, e.g. the polynucleotide encoding for the anti-SIRP α antibody or antigen-binding fragment thereof is introduced into the viral vector and then expressed in the body of the subject after infection with the virus.

[00249] In another aspect, expression of the anti-SIRP α antibody or antigen-binding fragment thereof is carried out in vertebrate cells. The propagation of vertebrate cells in culture (tissue culture) has become routine procedure and techniques are widely available. Examples of useful mammalian host cell lines are monkey kidney CV1 line transformed by SV40 (COS-7, ATCC CRL 1651), human embryonic kidney line (293 or 293 cells subcloned for growth in suspension culture, (Graham et al., 1977, J. Gen Virol. 36: 59), baby hamster kidney cells (BHK, ATCC CCL 10), Chinese hamster ovary cells/-DHFR1 (CHO, Urlaub et al., 1980, Proc. Natl. Acad. Sci. USA 77: 4216; e.g., DG44), mouse sertoli cells (TM4, Mather, 1980, Biol. Reprod. 23:243-251), monkey kidney cells (CV1 ATCC CCL 70), African green monkey kidney cells (VERO-76, ATCC CRL-1587), human cervical carcinoma cells (HELA, ATCC CCL 2), canine kidney cells

(MDCK, ATCC CCL 34), buffalo rat liver cells (BRL 3A, ATCC CRL 1442), human lung cells (W138, ATCC CCL 75), human liver cells (Hep G2, HB 8065), mouse mammary tumor (MMT 060562, ATCC CCL51), TR1 cells (Mather et al., 1982, Annals N.Y. Acad. Sci. 383: 44-68), MRC 5 cells, FS4 cells, and human hepatoma line (Hep G2).

[00250] Also included are nucleic acids that hybridize under low, moderate, and high stringency conditions, in particular under high stringency conditions, as defined herein, to all or a portion (e.g., the portion encoding the variable region) of the nucleotide sequence represented by isolated polynucleotide sequence(s) that encode an anti-SIRP α antibody or antibody fragment. The hybridizing portion of the hybridizing nucleic acid is typically at least 15 (e.g., 20, 25, 30 or 50) nucleotides in length. The hybridizing portion of the hybridizing nucleic acid is at least 80%, e.g., at least 90%, at least 95%, at least 98%, or at least 99% identical to the sequence of a portion or all of a nucleic acid encoding an anti-SIRP α polypeptide (e.g., a heavy chain or light chain variable region), or its complement. Hybridizing nucleic acids of the type described herein can be used, for example, as a cloning probe, a primer, e.g., a PCR primer, or a diagnostic probe. In one aspect, "high stringency conditions" means for probes of at least 100 nucleotides in length, prehybridization and hybridization at 42°C. in 5×SSPE, 0.3% SDS, 200 micrograms/ml sheared and denatured salmon sperm DNA, and 50% formamide, following standard Southern blotting procedures for 12 to 24 hours. The carrier material is finally washed three times each for 15 minutes using 0.2×SSC, 0.2% SDS at 65°C.

[00251] In one embodiment, the present invention relates to an isolated polynucleotide comprising a nucleotide sequence encoding a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 100, 101, 102, 103, 104, 105, 110, 111, 112, 113, 114, 115, 116, 117, 118, 119, 120, 121, 122, 123, 124, or 221.

[00252] In one embodiment, the present invention relates to an isolated polynucleotide comprising a nucleotide sequence encoding a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOs: 105, 106, 107, 108, 109, 125, 126, 109, 127, 128, 129, 130, or 222.

[00253] In one embodiment, the present invention relates to an isolated polynucleotide comprising a nucleotide sequence encoding a heavy chain region comprising the amino acid sequence of any one of SEQ NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 147, 148, 149, 150, 151, 152, 217, 135, 153, 154, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173, 219, 133, 134, 137, 132, or 136.

[00254] In one embodiment, the present invention relates to an isolated polynucleotide comprising the nucleotide sequence encoding of a light chain region comprising the amino acid sequence of any one any one of SEQ NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195, 218, 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216, 220, 176, 177, 180, 175, or 179.

Articles of Manufacture

[00255] In another aspect, an article of manufacture containing materials useful for the treatment of the disorders described above is included. The article of manufacture comprises a container and a label. Suitable containers include, for example, bottles, vials, syringes, and test tubes. The containers may be formed from a variety of materials such as glass or plastic. The container holds a composition that is effective for treating the condition and may have a sterile access port. For example, the container may be an intravenous solution bag or a vial having a stopper pierceable by a hypodermic injection needle. The active agent in the composition is the anti-SIRP α antibody or the antigen-binding fragment thereof. The label on or associated with the container indicates that the composition is used for treating the condition of choice. The article of manufacture may further comprise a second container comprising a pharmaceutically-acceptable buffer, such as phosphate-buffered saline, Ringer's solution, and dextrose solution. It may further include other materials desirable from a commercial and user standpoint, including other buffers, diluents, filters, needles, syringes, and package inserts with instructions for use.

EXAMPLES

[00256] The present disclosure is now described with reference to the following Examples. These Examples are provided for the purpose of illustration only and the disclosure should in no way be construed as being limited to these Examples, but rather should be construed to encompass any and all variations which become evident as a result of the teaching provided herein.

[00257] Without further description, it is believed that one of ordinary skill in the art can, using the preceding description and the following illustrative examples, make and utilize the compounds of the present disclosure and practice the claimed methods. The following working examples therefore, specifically point out the preferred embodiments of the present invention, and are not to be construed as limiting in any way the remainder of the disclosure.

Example 1: Generation of Anti-SIRP α antibodies

[00258] *Phage and yeast display.*

[00259] Initial efforts to generate anti-SIRP α antibodies of desired selectivity, i.e., being able to block hSIRP α V1 and hSIRP α V2 and lacking binding to hSIRP γ , are conducted using phage display. Biotinylated extracellular domains of hSIRP α V1, hSIRP α V2, and cynomologus SIRP α proteins are used as the target proteins in three sequential screening rounds. Unlabeled hSIRP γ extracellular domain is included in each screening round as a deselecting agent in order to remove antibodies that could bind to hSIRP γ extracellular domain.

[00260] Phage display yielded thirteen candidate antibodies that were further characterized. Four of the candidate antibodies could block hSIRP α V1 and hSIRP α V2, but each of these antibodies was determined to also bind to hSIRP γ . Nine of the candidate antibodies did not bind to hSIRP γ , but also could only block hSIRP α V1 or hSIRP α V2 but not both. Thus, it was determined that none of the thirteen candidate antibodies possessed the desired selectivity.

[00261] A similar effort to generate anti-SIRP α antibodies of desired selectivity is conducted using yeast display. The majority of the candidate antibodies identified by yeast display (59 out of 66) could block hSIRP α V1 and hSIRP α V2 but also bound to hSIRP γ . The remaining seven antibodies did not bind to hSIRP γ , but could only block hSIRP α V1 but not hSIRP α V2. Thus, similarly to the phage display, it was determined that none of the candidate antibodies possessed the desired selectivity.

[00262] *Immunization.*

[00263] AlivaMab kappa mice are immunized subcutaneously with combinations of recombinantly produced extracellular domains of human and cynomologus SIRP α proteins [NP 542970, CAA71403 and NP 001271679] following standard laboratory immunization techniques. Briefly, mice are immunized subcutaneously once per week for four weeks with protein and adjuvant mixtures. An aqueous adjuvant is used and is expected to improve generation of antibodies with the desired selectivity by decreasing the exposure of sites that are internal to the native protein conformation. After four immunizations, antibody titers are determined by ELISA methods. Animals that display a strong antibody response against all antigens are selected for antibody recovery.

[00264] *B-cell recovery and recombinant antibody production.*

[00265] Antigen-specific memory B-cell recovery is performed via single B-cell flow cytometry and cell sorting. Recovered antibody sequences are synthesized and screened for various characteristics, including but not limited to, affinity to SIRP α , binding to huSIRP α -V1 and huSIRP α -V2 CHO cell lines, and blocking of SIRP α :CD47 interaction. Representative antibodies exhibiting the desired properties are shown in Table 26-27: Antibody A, Antibody B, Antibody C, Antibody D, and Antibody E.

Example 2. Removal of sequence liabilities for Antibody A and Antibody E.

[00266] Fragment antibody (Fab) clones of the parent antibodies are prepared. Libraries are prepared with certain position variations so as to remove potential sequence liabilities (i.e. amino acid residues that may be immunogenic or that may create potential manufacturing problems). These mutated libraries usually have 5 to 10 binary positions per V-region. Separate saturation libraries are also made apart from single or double mutation libraries to address specific positions in more complex V-regions. Such libraries are prepared using standard methods known in the art and can be readily used by the skilled person.

[00267] The engineered Fabs are tested for desired properties, including affinity to huSIRP α -V1 and huSIRP α -V2. Representative variable regions for engineered antibodies for Antibody A and Antibody E are shown in Table 28-29.

[00268] **Example 3. Binding affinities (SPR K_D) of antibodies to purified recombinant human SIRP α , SIRP β 1, SIRP γ proteins**

[00269] The binding affinity of anti-SIRP α antibodies to the various SIRP α analytes is determined by surface plasmon resonance (SPR) using a ProteOn XPR36 (Bio-Rad). The running buffer for this experiment and all dilutions (except where stated) are done in 1 × HBS-EP+. The CM5 sensorchip is activated with equal mixture of EDC/NHS for 420 sec at a flow rate of 10 μ l/min and immobilized with Protein A/G beads (50 μ g/ml in 10 mM acetate pH 4.5) for 420 sec at a flowrate of 10 μ l/min resulting in ~ 2600-2900 RU of Protein A/G on the surface. The sensorchip is deactivated with 1M ethanolamine HCl for 420 sec at a flowrate of 10 μ l/min.

[00270] Each antibody is captured on the Protein A/G surface for 60 sec at a flowrate of 10 μ l/min resulting in capture levels of ~280 RU. The analyte is injected over the captured

antibody for 300 sec at a flowrate of 30 μ l/min. The dissociation is done for 600 sec. Two-fold serial dilutions of each SIRP protein (Extracellular Domain + His-Tag) are injected as analyte with the highest concentrations indicated in table 36 below.

Table 36: List of various SIRP proteins and their highest concentration used for binding affinity analysis

Analyte (Extracellular Domain + His-Tag)	Accession Number	Highest Concentration (nM) in experiment
hSIRP α V1	NP_542970.1	100
hSIRP α V2	CAA71403.1	100
hSIRP β 1	NP_006056.2	1000
hSIRP β 1v3	NP_001129316.1	100
hSIRP γ	NP_061026.2	1000
cynoSIRP α	NP_001271679	1000
cynoSIRP α	EGM_02252	1000
cynoSIRP α	XP_015313155	1000
cynoSIRP β 1	XP_005568598	1000
cynoSIRP β 1v3	XP_005568593	1000
cynoSIRP γ	XP_005568591	1000

[00271] Once each analyte injection is complete, the sensor surface is regenerated by injecting 0.85% phosphoric acid for 30 sec at a flowrate of 30 μ l/min.

[00272] The analyte interaction with sensor surface (flow cell 1) and blank (HBS-EP+) are subtracted from the raw data. Sensorgrams are then fit globally to 1:1 Langmuir binding to provide on-rate (k_a), off-rate (k_D), and affinity (K_D) values as well as the steady state binding mode to provide the equilibrium affinity (K_D).

[00273] The above procedure is used to measure the binding affinity to the following analytes: human SIRP α V1, human SIRP α V2, human SIRP β 1, human SIRP β 1v3, human SIRP γ , cynomolgus (cyno) SIRP α , cyno SIRP β 1, cyno SIRP β 1v3, cyno SIRP γ , mouse SIRP α . The results for binding affinities of SIRP α antibodies are shown in Tables 37-42 below.

Table 37. Binding affinity of SIRP α antibodies (Antibodies A-G) to human SIRP proteins.

Antibodies	HuSIRP α V1 (M)	HuSIRP α V2 (M)	HuSIRP β 1 (M)	HuSIRP β 1v3 (M)	HuSIRP γ (M)
Antibody A	8.81E-09	1.31E-08	7.82E-08	NB	NB
Antibody B	1.74E-08	7.15E-08	NB	NB	NB
Antibody C	6.27E-10	9.69E-11	2.14E-09	NB	NB
Antibody D	4.04E-09	4.05E-09	1.65E-08	NB	NB
Antibody E	4.56E-10	4.2E-10	1.23E-06	1.16E-09	NB
Antibody F	9.30E-09	1.41E-08	8.40E-08	NB	NB
Antibody G	4.97E-10	4.65E-10	1.27E-06	1.27E-09	NB

NB=No binding at highest concentration

Table 38. Binding affinity of SIRP α antibodies (Antibodies A-G) to cyno and mouse SIRP proteins.

Antibodies	Mouse NP_031573 SIRP α (M)	CySIRP α NP_001271679 (M)	CySIRP α EGM_02252 (M)	CySIRP α XP_005568593 (M)
Antibody A	NB	4.53E-07	2.01E-07	2.01E-07
Antibody B	NB	NB	NA	NA
Antibody C	NB	1.43E-08	NA	NA
Antibody D	NB	NB	NA	NA
Antibody E	NB	9.21E-09	NB	6.98E-09
Antibody F	NB	5.06E-07	2.30E-07	2.27E-07
Antibody G	NB	1.01E-08	NB	7.62E-09

NB=No binding at highest concentration

NA=Data Not Available

Table 39. Binding affinity of SIRP α antibodies (Antibodies A, A1-A16) to human SIRP proteins.

Antibodies	HuSIRP α V1 (M)	HuSIRP α V2 (M)	HuSIRP β 1 (M)	HuSIRP β 1v3 (M)	HuSIRP γ (M)
Antibody A	7.72E-09	9.03E-09	6.48E-08	NB	NB
Antibody A15	7.06E-09	7.91E-09	5.88E-08	NB	NB
Antibody A13	7.28E-09	8.36E-09	6.10E-08	NB	NB
Antibody A9	7.34E-09	8.25E-09	5.88E-08	NB	NB
Antibody A12	7.83E-09	8.96E-09	6.44E-08	NB	NB
Antibody A10	7.95E-09	9.17E-09	6.90E-08	NB	NB
Antibody A16	7.98E-09	8.22E-09	6.27E-08	NB	NB
Antibody A14	8.13E-09	7.95E-09	5.83E-08	NB	NB
Antibody A6	8.28E-09	1.81E-08	6.51E-08	NB	NB
Antibody A4	8.46E-09	1.83E-08	6.72E-08	NB	NB

Antibody A8	8.53E-09	1.88E-08	6.96E-08	NB	NB
Antibody A1	8.60E-09	1.86E-08	NB	NB	NB
Antibody A5	8.75E-09	1.78E-08	6.42E-08	NB	NB
Antibody A3	9.06E-09	1.96E-08	7.03E-08	NB	NB
Antibody A7	9.38E-09	2.04E-08	7.22E-08	NB	NB
Antibody A2	9.39E-09	2.09E-08	7.75E-08	NB	NB

NB=No binding at highest concentration

Table 40. Binding affinity of SIRP α antibodies (Antibodies A, A1-A16) to cyno SIRP proteins.

Antibodies	CySIRPα NP_001271679 (M)	CySIRPα EGM_02 252 (M)	CySIRPα XP_01531315 5 (M)	Cyno SIRPβ1 XP_005 568598 (M)	Cyno SIRPβ1v3 XP_005568 593 (M)	Cyno SIRPγ XP_00556859 1(M)
Antibody A	3.36E-07	2.58E-07	1.65E-07	NB	2.46E-07	NB
Antibody A15	3.17E-07	2.23E-07	1.54E-07	NB	2.32E-07	NB
Antibody A13	3.30E-07	2.26E-07	1.60E-07	NB	2.38E-07	NB
Antibody A9	3.46E-07	2.27E-07	1.56E-07	NB	2.57E-07	NB
Antibody A12	3.84E-07	2.49E-07	1.82E-07	NB	2.79E-07	NB
Antibody A10	3.78E-07	2.49E-07	1.80E-07	NB	2.71E-07	NB
Antibody A16	3.06E-07	2.25E-07	1.51E-07	NB	2.25E-07	NB
Antibody A14	3.16E-07	2.22E-07	1.53E-07	NB	2.35E-07	NB
Antibody A6	6.84E-07	5.38E-07	3.66E-07	NB	4.69E-07	NB
Antibody A4	6.69E-07	5.08E-07	3.74E-07	NB	4.60E-07	NB
Antibody A8	6.39E-07	5.35E-07	3.50E-07	NB	4.49E-07	NB
Antibody A1	7.80E-07	5.43E-07	3.69E-07	NB	5.57E-07	NB
Antibody A5	6.65E-07	5.33E-07	3.47E-07	NB	4.65E-07	NB
Antibody A3	8.57E-07	5.71E-07	4.38E-07	NB	5.64E-07	NB
Antibody A7	7.41E-07	6.09E-07	3.89E-07	NB	4.94E-07	NB

Antibody A2	8.87E-07	5.84E-07	4.24E-07	NB	5.52E-07	NB
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NB=No binding at highest concentration

NA=Data Not Available

Table 41. Binding affinity of SIRP α antibodies (Antibodies E, E1-E22) to human SIRP proteins.

Antibodies	HuSIRPα V1 (M)	huSIRPαV2 (M)	huSIRPβ1 (M)	huSIRPβ1v3 (M)	huSIRPγ (M)
Antibody E	4.25E-10	3.30E-10	7.71E-07	8.10E-10	NB
Antibody E1	7.23E-10	6.30E-10	1.48E-06	1.81E-09	NB
Antibody E2	9.12E-10	6.87E-10	NB	1.93E-09	NB
Antibody E3	9.64E-10	7.75E-10	2.14E-06	1.76E-09	NB
Antibody E4	9.88E-10	8.24E-10	1.34E-06	1.95E-09	NB
Antibody E5	1.02E-09	8.73E-10	1.38E-06	2.72E-09	NB
Antibody E6	1.03E-09	8.75E-10	NB	2.72E-09	NB
Antibody E7	1.20E-09	1.18E-09	8.74E-07	2.21E-09	NB
Antibody E8	1.21E-09	5.42E-10	2.42E-06	2.58E-09	NB
Antibody E9	1.22E-09	9.41E-10	NB	3.16E-09	NB
Antibody E10	1.23E-09	1.20E-09	1.57E-06	2.37E-09	NB
Antibody E11	1.41E-09	5.68E-10	3.28E-06	2.99E-09	NB
Antibody E12	1.67E-09	1.26E-09	1.67E-06	3.43E-09	NB
Antibody E13	1.70E-09	6.90E-10	NB	4.17E-09	NB
Antibody E14	1.84E-09	1.36E-09	NB	4.80E-09	NB
Antibody E15	1.91E-09	7.37E-10	NB	4.56E-09	NB
Antibody E16	2.15E-09	1.93E-09	4.24E-06	4.02E-09	NB
Antibody E17	2.36E-09	7.97E-10	NB	5.32E-09	NB
Antibody E18	2.46E-09	8.98E-10	NB	4.80E-09	NB
Antibody E19	2.52E-09	9.70E-10	1.66E-06	4.74E-09	NB
Antibody E20	3.42E-09	1.11E-09	4.35E-07	8.48E-09	NB
Antibody E21	4.46E-09	1.39E-09	NB	8.84E-09	NB
Antibody E22	1.57E-09	1.47E-09	NB	3.19E-09	NB

NB=No binding at highest concentration

Table 42. Binding affinity of Antibody E, E1-E22 SIRP α antibodies to cyno SIRP proteins.

Antibodies	CySIRP α NP_001271679 (M)	CySIRP α EGM_022 52 (M)	CySIRP α XP_015313155 (M)	Cyno SIRP β 1 XP_0055685 98 (M)	Cyno SIRP β 1v 3 XP_0055 68593 (M)	Cyno SIRP γ XP_005568 591 (M)
Antibody E	7.46E-09	NB	7.07E-09	NB	4.62E-09	NB
Antibody E1	1.79E-08	NB	1.67E-08	NB	1.08E-08	NB
Antibody E2	2.07E-08	NB	1.97E-08	NB	1.24E-08	NB
Antibody E3	1.73E-08	NB	1.69E-08	NB	1.07E-08	NB
Antibody E4	2.11E-08	NB	1.98E-08	NB	1.31E-08	NB
Antibody E5	3.82E-08	NB	3.78E-08	NB	2.26E-08	NB
Antibody E6	4.10E-08	NB	4.16E-08	NB	2.39E-08	NB
Antibody E7	2.19E-08	NB	2.12E-08	NB	1.31E-08	NB
Antibody E8	1.77E-08	NB	1.66E-08	NB	1.08E-08	NB
Antibody E9	4.37E-08	NB	4.08E-08	NB	2.51E-08	NB
Antibody E10	2.52E-08	NB	2.37E-08	NB	1.57E-08	NB
Antibody E11	2.03E-08	NB	2.00E-08	NB	1.32E-08	NB
Antibody E12	4.61E-08	NB	4.22E-08	NB	2.79E-08	NB
Antibody E13	3.83E-08	NB	3.74E-08	NB	2.05E-08	NB
Antibody E14	9.07E-08	NB	8.63E-08	NB	5.19E-08	NB
Antibody E15	4.94E-08	NB	4.62E-08	NB	2.58E-08	NB
Antibody E16	5.03E-08	NB	4.88E-08	NB	3.03E-08	NB
Antibody E17	4.48E-08	NB	4.32E-08	NB	2.70E-08	NB
Antibody E18	5.25E-08	NB	5.00E-08	NB	2.78E-08	NB

Antibody E19	4.66E-08	NB	4.47E-08	NB	2.50E-08	NB
Antibody E20	1.11E-07	NB	1.04E-07	NB	5.74E-08	NB
Antibody E21	1.07E-07	NB	1.08E-07	NB	5.65E-08	NB
Antibody E22	1.38E-08	NB	1.42E-08	NB	1.20E-08	NB

NB=No binding at highest concentration

Example 4. Binding of antibodies to full-length human V1-SIRPα or V2-SIRPα expressed on CHO cells

[00274] Antibody binding to cells expressing SIRPα is evaluated by flow cytometry. CHO-K1 parental cells (negative control), CHO-K1 cells expressing full-length human SIRPαV1 (NP_542970.1) or CHO-K1 cells expressing full-length human SIRPαV2 (CAA71403.1) are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00275] Anti-SIRPα antibody KWAR23 (WO2015138600) and antibodies A-E demonstrated dose-dependent binding to CHO cells expressing full-length human SIRPαV1 (Figure 1B) or SIRPαV2 (Figure 1C) with EC₅₀ values ranging between 0.4-14 nM (Table 43). None of the antibodies tested exhibited binding to parental CHO cells (Figure 1A).

Table 43: EC₅₀ of Anti-SIRPα antibodies

Antibody	Cell Binding EC ₅₀ (nM)		
	CHO-Parental	CHO-hSIRPαV1	CHO-hSIRPαV2
Isotype	NB	NB	NB
KWAR23	NB	1.0	0.5
Antibody A	NB	0.9	0.4
Antibody B	NB	12.0	14.2
Antibody C	NB	1.0	0.5
Antibody D	NB	0.7	0.5
Antibody E	NB	2.0	0.9

^{NB}No binding**Example 5. Binding of antibodies to endogenous human V1-SIRPα or V2-SIRPα expressed on human monocytic cell lines**

[00276] Antibody binding to human monocytic cell lines homozygous for the V1-allele (U-937) or the V2-allele (THP-1) of SIRPα is assessed. U-937 or THP-1 cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00277] Anti-SIRPα antibody KWAR23 and antibodies A-E demonstrated dose-dependent binding to U-937 cells homozygous for human SIRPαV1 allele (Figure 2A) and THP-1 cells homozygous for human SIRPαV2 allele (Figure 2B) with EC₅₀ values ranging between 0.2-22 nM (Table 44).

Table 44: EC₅₀ of Anti-SIRPα antibodies

Antibody	Cell Binding EC ₅₀ (nM)	
	U937	THP-1
Isotype	NB	NB
KWAR23	0.5	0.2
Antibody A	0.4	0.2
Antibody B	21.1	22.3
Antibody C	0.6	0.3
Antibody D	0.5	0.3
Antibody E	0.5	0.4

^{NB}No Binding

Example 6. Binding of antibodies to endogenous human V1-SIRP α and/or V2-SIRP α expressed on primary human monocytes

[00278] Antibody binding to primary human cells expressing SIRP α is evaluated by flow cytometry. Human peripheral blood mononuclear cells (PBMCs) from donors genotyped as homozygous for the SIRP α V1 allele, V2 allele, or heterozygous for both alleles are blocked with donkey IgG and incubated on ice with increasing concentrations of antibody for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Washed cells are subsequently blocked with Human TruStain FcX™ (Biolegend) and stained with BV421-conjugated anti-human CD14 (Biolegend, clone M5E2). Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity of the CD14+ population was determined and used as a measure of antibody binding.

[00279] All antibodies tested (Antibody A-G, A10, A4, A11, and E22) demonstrated dose-dependent binding to primary human monocytes from donors homozygous for V1 (Figures 3A-3D) or V2 (Figures 5A-5D) alleles or heterozygous for both alleles (Figures 4A-4D). Table 45 highlights the primary cell binding EC₅₀ calculated for each of the antibodies described above. Aside from Antibody B, all antibodies exhibited subnanomolar EC₅₀.

[00280] Binding of known anti-SIRP α antibodies, KWAR23, 1H9 (WO 2019/023347) and SIRP AB11 (WO 2020/068752), to primary human monocytes were also assessed as described above and EC₅₀ results are shown in Table 45. While 1H9 exhibited dose-dependent binding and subnanomolar binding EC₅₀ values for primary human monocytes from donors homozygous for V1 alleles or heterozygous for both alleles (Figures 3C and 4C), non-saturable antibody binding was detected on primary human monocytes from donors homozygous for V2 alleles. KWAR23 and SIRP AB11 antibodies exhibited subnanomolar binding EC₅₀ on primary human monocytes from donors of all genotypes (Figures 3A-3D, 4A-4D and 5A-5D; Table 45).

Table 45: EC₅₀ of Anti-SIRP α antibodies

Antibody	Primary Human Monocyte Cell Binding EC ₅₀ ; nM $\pm$ SD (n)		
	V1V1	V1V2	V2V2
Isotype	NB	NB	NB
KWAR23	0.2 $\pm$ 0.04 (n=7)	0.1 $\pm$ 0.1 (n=7)	0.1 $\pm$ 0.05 (n=7)
Antibody A	0.2 $\pm$ 0.03 (n=5)	0.2 $\pm$ 0.1 (n=5)	0.1 $\pm$ 0.04 (n=5)
Antibody B	26.6 (n=1)	24.1 (n=1)	21.8 (n=1)
Antibody C	0.6 (n=1)	0.5 (n=1)	0.3 (n=1)
Antibody D	0.3 (n=1)	0.5 (n=1)	0.2 (n=1)
Antibody E	0.4 $\pm$ 0.1 (n=5)	0.4 $\pm$ 0.2 (n=5)	0.7 $\pm$ 0.3 (n=5)

Antibody F	0.2 (n=1)	0.1 (n=1)	0.2 (n=1)
Antibody G	0.3 (n=1)	0.3 (n=1)	0.4 (n=1)
Antibody A10	0.2 ± 0.04 (n=5)	0.1 ± 0.05 (n=5)	0.1 ± 0.04 (n=5)
Antibody A4	0.1 ± 0.01 (n=3)	0.1 ± 0.04 (n=3)	0.1 ± 0.03 (n=3)
Antibody E22	0.8 ± 0.1 (n=3)	0.7 ± 0.2 (n=3)	0.4 ± 0.2 (n=3)
Antibody A11	0.1 ± 0.04 (n=2)	0.1 ± 0.06 (n=2)	0.1 ± 0.07 (n=2)
1H9	0.2 (n=1)	0.2 (n=1)	NS (n=1)
SIRP AB11	0.1 ± 0.01 (n=2)	0.1 ± 0.01 (n=2)	0.1 ± 0.001 (n=2)

^{NB}No Binding

^{NS}Non-saturable binding

Example 7. Binding of antibodies to full-length human SIRPβ1 or SIRPβL-expressed on U937^{SIRPα KO} cells

[00281] Antibody binding to cells expressing full-length human SIRPβ1 (NP_006056.2) and SIRPβL (NP_001129316.1) is evaluated by flow cytometry. SIRPβ1- or SIRPβL-expressing U937^{SIRPα KO} cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00282] Antibodies A-G, A10, A4, and A11, but not isotype control or antibody E22, demonstrated dose-dependent binding to full length human SIRPβ1 expressed on U-937^{SIRPα KO} cells (Figures 6A-6D). Table 44 highlights the binding EC₅₀ calculated for each of the antibodies described above. While antibodies A, C, D, F, A10, A4, and A11 exhibited subnanomolar EC₅₀ values to these cells, binding of antibodies B, E, and G were non-saturable at concentrations as high as 200 nM and no EC₅₀ values determined.

[00283] Antibodies E, G, and E22 demonstrated dose-dependent binding to full length human SIRPβL expressed on U-937^{SIRPα KO} cells (Figures 7A-7D) with EC₅₀ less than 1 nM (Table 46). No binding of antibodies A, C, D, F, A10, A4 and A11 could be detected on SIRPβL-expressing U-937^{SIRPα KO} cells (Figures 7A-7D). Binding of antibody B (Figure 7A) was detected on SIRPβL-expressing U-937^{SIRPα KO} cells; however, binding was non-saturable at concentrations as high as 200 nM

[00284] Binding of known anti-SIRPα antibodies, KWAR23, 1H9 and SIRPAB11, to SIRPβ1- or SIRPβL-expressing U937^{SIRPα KO} cells was also assessed as described above. All 3 molecules demonstrated dose-dependent binding to both cell types (Figures 6A-6D and 7A-7D) with sub-nanomolar binding EC₅₀ values (Table 46).

Table 46: EC₅₀ of Anti-SIRPα antibodies

Antibody	Cell Binding EC ₅₀ ; nM ± SD (n)	
	SIRPβ1	SIRPβL
Isotype	NB (n=6)	NB (n=6)
KWAR23	0.2 ± 0.1 (n=6)	0.2 ± 0.1 (n=6)
Antibody A	0.2 ± 0.1 (n=5)	NB (n=5)
Antibody B	NS (n=1)	NS (n=1)
Antibody C	0.5 (n=1)	NB (n=1)
Antibody D	0.4 (n=1)	NB (n=1)
Antibody E	NS (n=5)	0.3 ± 0.1 (n=5)
Antibody F	0.2 (n=1)	NB (n=1)
Antibody G	NS (n=1)	0.3 (n=1)
Antibody A10	0.2 ± 0.1 (n=4)	NB (n=4)
Antibody A4	0.2 ± 0.1 (n=3)	NB (n=3)
Antibody E22	NB (n=3)	0.6 ± 0.2 (n=3)
Antibody A11	0.3 (n=1)	NB (n=1)
1H9	0.2 ± 1.0 (n=2)	0.3 ± 0.2 (n=2)
SIRPAB11	0.1 (n=1)	0.2 (n=1)

^{NB}No binding^{NS}Non-saturable binding as high as 200 nM**Example 8. Binding of antibodies to purified recombinant human SIRPγ proteins by ELISA**

[00285] A biochemical ELISA-based assay is utilized to evaluate the binding of antibodies to SIRPγ. Black, 96-well flat-bottom immunoassay plates are coated with recombinant human SIRPγ extracellular domain (ECD)/His protein (NP_061026.2) diluted in DPBS and incubated overnight at 4°C. Plates are washed three times and subsequently blocked for about 1-2h at room temperature (RT). Plates are washed and serially titrated antibodies are added to respective wells for an incubation of about 1-2h at RT. After the incubation, the plates are washed and HRPO-conjugated mouse anti-human IgG secondary reagent (Jackson Immuno Research) is added to each well and incubated for about 1-2 hours. Following the final plate washing, Amplex™ red substrate is added and incubated at RT in the dark for 15-30 minutes. Fluorescence is detected ($\lambda_{Ex}/\lambda_{Em}$ = 530/590 nm) using EnVision Multilabel Plate Reader (Perkin Elmer). Average fluorescence is plotted for measuring the antibody binding.

[00286] Except SIRPγ-reactive mAb, KWAR23, none of the antibodies tested (Antibodies A-E) exhibited binding to immobilized recombinant human SIRPγ ECD/His protein (Figure 8). The EC₅₀ value for KWAR23 was determined to be 0.1 nM (Table 47).

Example 9. Binding of antibodies to full-length human SIRP γ expressed on CHO cells

[00287] Antibody binding to cells expressing human SIRP γ (NP_061026.2) is evaluated by flow cytometry. SIRP γ -expressing CHO cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibody for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00288] Binding for each of the antibodies on SIRP γ -expressing CHO cells was detected relative to isotype control (Figure 9). Compared to SIRP γ -reactive mAb (KWAR23) that exhibited an EC₅₀ of 2.7 nM (Table 47), binding of antibodies A and C-E was non-saturable at concentrations as high as 200 nM and the magnitude of bound antibody detected was low.

Example 10. Binding of antibodies to SIRP γ expressed on primary human T cells

[00289] Antibody binding to primary human T cells expressing endogenous SIRP γ is evaluated by flow cytometry. Human peripheral blood mononuclear cells (PBMCs) are blocked with donkey IgG and incubated on ice with increasing concentrations of antibody for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Human PBMCs are subsequently blocked with Human TruStain FcX™ (Biolegend) and stained with BUV395-conjugated anti-human CD3 (BD Biosciences, clone SK7). Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity of CD3+ gated population is determined and used as a measure of antibody binding.

[00290] Binding of known anti-SIRP α antibodies, KWAR23, 1H9 and SIRPAB11, to primary human T cells is also assessed as described above. KWAR23, 1H9 and SIRP AB11 demonstrated dose-dependent binding to primary human CD3+ cells (Figures 10A-10D) with EC₅₀ values less than 1 nM (Table 47), while no binding by antibodies A-G, A10, A4, A11, and E22 was detected.

Table 47: EC₅₀ of Anti-SIRPα antibodies

Antibody	EC ₅₀ , nM ± SD (n)		
	recombinant hSIRPγ	CHO-hSIRPγ	primary CD3+cells
Isotype	NB (n=1)	NB (n=1)	NB (n=18)
KWAR23	0.1 (n=1)	2.7 (n=1)	0.03 ± 0.01 (n=15)
Antibody A	NB (n=1)	NS (n=1)	NB (n=15)
Antibody B	NB (n=1)	11.8 (n=1)	NB (n=3)
Antibody C	NB (n=1)	NS (n=1)	NB (n=3)
Antibody D	NB (n=1)	NS (n=1)	NB (n=3)
Antibody E	NB (n=1)	NS (n=1)	NB (n=15)
Antibody F	NT	NT	NB (n=3)
Antibody G	NT	NT	NB (n=3)
Antibody A10	NT	NT	NB (n=12)
Antibody A4	NT	NT	NB (n=9)
Antibody E22	NT	NT	NB (n=9)
Antibody A11	NT	NT	NB (n=3)
1H9	NT	NT	0.2 ± 0.1 (n=2)
SIRPAB11	NT	NT	0.02 ± 0.004 (n=3)

^{NB}No binding^{NT}Not tested^{NS}Non-saturable binding

Example 11. Blocking of human CD47 binding to full-length human V1-SIRPα or V2-SIRPα expressed on CHO cells

[00291] Human CD47 tetramers are assembled using biotinylated human CD47 (NP_942088; AcroBiosystems) and AF647-conjugated streptavidin (Biolegend). To assess antibody ligand blocking potential, CHO-K1 cells expressing full-length human SIRPαV1 (NP_542970.1) or CHO-K1 cells expressing full-length human SIRPαV2 (CAA71403.1) are co-incubated with increasing concentrations of antibody and a fixed concentration of human CD47 tetramer on ice for 60 min. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of CD47 binding.

[00292] Known KWAR23 antibody and antibodies A-E dose-dependently blocked human CD47 binding to human SIRPαV1 and SIRPαV2-expressing CHO cells (Figures 11A and 11B) with half maximal inhibitory concentration (IC₅₀) values ranging between 0.3-6.0 nM (Table 48). Blockade of human CD47 binding to human SIRPαV2-expressing CHO cells by antibody E plateaued at about 80%.

Table 48: IC₅₀ of Anti-SIRP α antibodies

Antibody	Blockade of Human CD47 Binding; IC ₅₀ (nM)	
	CHO-hSIRP α V1	CHO-hSIRP α V2
Isotype	NB	NB
KWAR23	0.9	0.6
Antibody A	0.6	0.3
Antibody B	5.6	6.0
Antibody C	0.7	0.3
Antibody D	0.6	0.3
Antibody E	0.7	0.7*

^{NB}No blockade

*Inhibition plateaus at <100%

Example 12. Blocking of human CD47 binding to endogenous human V1-SIRP α and/or V2-SIRP α expressed on primary human monocytes

[00293] Human CD47 tetramers are assembled using biotinylated human CD47 (NP_942088; AcroBiosystems) and AF647-conjugated streptavidin (Biolegend). Human peripheral blood mononuclear cells (PBMCs) from donors genotyped as homozygous for the SIRP α V1 allele, V2 allele, or heterozygous for both alleles are blocked with Human TruStain FcX™ (Biolegend) and co-incubated with increasing concentrations of antibody, a fixed concentration of human CD47 tetramer and BV421-conjugated anti-human CD14 on ice for 60 min. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity of CD14+ gated population is determined and used as a measure of CD47 binding.

[00294] Antibodies A-G, A10, A4, E22, and A11, but not isotype control, dose-dependently blocked human CD47 binding to primary human monocytes from donors homozygous for V1 (Figures 12A-12D) or V2 (Figures 14A-14D) alleles or heterozygous for both alleles (Figures 13A-13D). Aside from Antibody B, all antibodies exhibited IC₅₀ values less than 1 nM (Table 49).

[00295] The ability of known anti-SIRP α antibodies, KWAR23, 1H9 and SIRPAB11, to block human CD47 binding to primary human monocytes is assessed as described above. While 1H9 blocked human CD47 from binding to primary human monocytes from donors homozygous for V1 alleles (Figure 12C) or heterozygous for both alleles (Figure 13C) with subnanomolar IC₅₀ values (Table 49), incomplete blockade to primary human monocytes from donors homozygous for V2 alleles was observed (Figure 14C). KWAR23 and SIRPAB11 antibodies blocked human

CD47 binding to primary human monocytes from donors of all genotypes (Figures 12A-12D, 13A-13D and 14A-14D; Table 49).

Table 49: IC₅₀ of Anti-SIRPα antibodies

Antibody	Blockade of Human CD47 Binding; IC ₅₀ , nM ± SD (n)		
	V1V1	V1V2	V2V2
Isotype	NB (n=6)	NB (n=6)	NB (n=6)
KWAR23	0.07 ± 0.04 (n=6)	0.10 ± 0.06 (n=6)	0.11 ± 0.06 (n=6)
Antibody A	0.06 ± 0.03 (n=5)	0.07 ± 0.05 (n=5)	0.06 ± 0.03 (n=5)
Antibody B	2.55 (n=1)	2.77 (n=1)	4.60 (n=1)
Antibody C	0.04 (n=1)	0.04 (n=1)	0.05 (n=1)
Antibody D	0.03 (n=1)	0.04 (n=1)	0.05 (n=1)
Antibody E	0.09 ± 0.03 (n=5)	0.10 ± 0.04 (n=5)	0.11 ± 0.04 (n=5)
Antibody F	0.07 (n=1)	0.07 (n=1)	0.11 (n=1)
Antibody G	0.11 (n=1)	0.11 (n=1)	0.22 (n=1)
Antibody A10	0.05 ± 0.01 (n=4)	0.07 ± 0.08 (n=4)	0.05 ± 0.02 (n=4)
Antibody A4	0.04 ± 0.01 (n=3)	0.05 ± 0.04 (n=3)	0.07 ± 0.04 (n=3)
Antibody E22	0.25 ± 0.10 (n=3)	0.36 ± 0.21 (n=3)	0.30 ± 0.10 (n=3)
Antibody A11	0.03 (n=1)	0.03 (n=1)	0.05 (n=1)
1H9	0.04 (n=1)	0.15 (n=1)	NS (n=1)
SIRPAB11	0.02 (n=1)	0.05 (n=1)	0.05 (n=1)

^{NB}No Blockade

^{NS}Non-saturable inhibition

Example 13. Blocking human V1-SIRPα and V2-SIRPα signaling dependent on cellular CD47 expression

[00296] A SIRPα signaling assay is used to measure SIRPα engagement induced by CD47 presented via cell-cell interaction. Detection of SIRPα signaling in this assay relies on enzyme fragment complementation (EFC). EFC uses a complementation system in β-galactosidase, which consists of the enzyme donor (ED) and the enzyme acceptor (EA) fragments. Independently, these fragments have no β-gal enzymatic activity; however, when brought into proximity they form an active β-gal enzyme. The reporter Jurkat cells are devoid of CD47 and stably co-express an ED-tagged SIRPα receptor and EA-tagged SH2 domain of the SHP-1 phosphatase. When the reporter cells are exposed to donor cells (Jurkat cells expressing endogenous levels of CD47), SIRPα is phosphorylated and recruits SHP-1 phosphatase. This interaction forces ED and EA fragments complementation and formation of active β-gal enzyme that is capable of hydrolyzing substrate to generate a chemiluminescent signal as a measure of receptor activation.

[00297] Jurkat SIRP α -V1 or - α V2 signaling cells (20,000) are incubated with serially titrated antibodies in 96-well white-bottom TC treated plates for 1h at 37°C, 5% CO₂ prior to the addition of Jurkat^{Parental} cells (30,000) in a total volume of 100 μ l in PathHunter cell plating 0 reagent. Plates are incubated in a humidified incubator for 5 hours at 37°C, 5% CO₂. Following the addition of PathHunter BioAssay Detection Reagents and incubation at room temperature in the dark, the plates are read on an EnVision® luminometer. Average RLU units are plotted as a measure of receptor activation.

[00298] Antibodies A-G, A10, A4, E22, and A11, but not isotype control, completely blocked cellular CD47-mediated SIRP α V1 activation (Figures 15A-15D) with subnanomolar IC₅₀ values (Table 48). Similarly, all antibodies dose-dependently blocked cellular CD47-mediated SIRP α V2 activation (Figures 16A-16D; Table 50), though inhibition by antibodies E and E22 plateaued at about 80% and 55%, respectively and antibody B did not achieve saturable inhibition.

[00299] The ability of known SIRP α antibodies, KWAR23, 1H9 and SIRPAB11, to block cellular CD47-mediated SIRP α signaling is assessed as described above. While 1H9 completely blocked cellular CD47-mediated SIRP α V1 activation with an IC₅₀ of 0.04 nM (Figure 15C; Table 50), blockade of cellular CD47-mediated SIRP α V2 activation did not achieve saturable inhibition (Figure 16C; Table 50). KWAR23 and SIRP AB11 antibodies were capable of blocking cellular CD47-mediated SIRP α signaling of by cells expressing either SIRP α allele.

Table 50: IC₅₀ of Anti-SIRP α antibodies

Antibody	Blockade of Cellular CD47-Mediated SIRP α Signaling; IC ₅₀ ; nM $\pm$ SD (n)	
	V1V1	V2V2
Isotype	NB (n=6)	NB (n=6)
KWAR23	0.04 $\pm$ 0.01 (n=6)	0.04 $\pm$ 0.01 (n=6)
Antibody A	0.04 $\pm$ 0.01 (n=5)	0.04 $\pm$ 0.01 (n=5)
Antibody B	0.76 (n=1)	NS (n=6)
Antibody C	0.04 (n=1)	0.03 (n=1)
Antibody D	0.04 (n=1)	0.04 (n=1)
Antibody E	0.04 $\pm$ 0.01 (n=5)	0.04 $\pm$ 0.01 (n=5)*
Antibody F	0.04 (n=1)	0.03 (n=1)
Antibody G	0.04 (n=1)	0.03 (n=1)
Antibody A10	0.04 $\pm$ 0.01 (n=4)	0.04 $\pm$ 0.01 (n=4)
Antibody A4	0.04 $\pm$ 0.01 (n=3)	0.05 $\pm$ 0.01 (n=3)
Antibody E22	0.07 $\pm$ 0.01 (n=3)	0.07 $\pm$ 0.01 (n=3)*
Antibody A11	0.04 (n=1)	0.02 (n=1)

1H9	0.04 (n=1)	NS (n=1)
SIRPAB11	0.04 (n=1)	0.03 (n=1)

^{NB}No Blockade
^{NS}Non-saturable inhibition
 *Inhibition plateaus at <100%

Example 14. SIRPα antagonist revert inhibition of phagocytosis caused by CD47 in human monocytic cell lines

[00300] A flow-based phagocytosis assay was developed to quantify the SIRPα-mediated regulation of phagocytosis induced by CD47. U-937 cells devoid of endogenous SIRPα are transduced with lentivirus containing full-length V1- or V2-SIRPα alleles. These cells are preincubated with anti-SIRPα antibodies at the indicated concentrations in 96-well V-bottom tissue culture (TC)-treated plates for 30 minutes prior to the addition of fluorescently conjugated microbeads (Spherotech) coated with or without human CD47 (NP_942088; AcroBiosystems) and opsonized with human IgG1 antibodies to induce Fc-receptor mediated phagocytosis. Following a 2h incubation at 37°C, 5% CO₂, samples are washed, blocked with Human TruStain FcX™ (Biolegend) and stained with fluorescently-conjugated antibody that recognize the external surface of the beads to enable differentiation between beads stuck on the outside of the cells and those that were inside. Cells are washed, fixed, and analyzed by flow cytometry. Phagocytosis is represented as the percentage of cells with internalized beads.

[00301] Internalization of opsonized beads by U-937 cells expressing SIRPα for either V1 or V2 alleles dropped when cells were incubated with CD47-coated beads (white bars); an effect not observed when cells were incubated with beads lacking CD47 (black bars). Pre-treatment of cells with 67 nM of the indicated antibodies restored both V1- (Figure 17A) and V2- (Figure 17B) SIRPα-expressing U-937 cell's ability to phagocytize CD47-coated beads (white bars) indicating that the antibodies can block CD47-mediated SIRPα signaling.

Example 15. SIRPα antagonist revert inhibition of phagocytosis caused by CD47 in primary human monocyte-derived macrophages of any combination of SIRPα alleles

[00302] A flow-based phagocytosis assay was developed to quantify the SIRPα-mediated regulation of phagocytosis induced by CD47. Monocytes enriched from human PBMCs (EasySep Monocyte Enrichment Kit, StemCell Technologies) from donors genotyped as homozygous for the SIRPα V1 allele, V2 allele, or heterozygous for both alleles are differentiated into macrophages following 6-7 days of culture in ImmunoCult™-SF macrophage differentiation

medium spiked with 50 ng/ml human recombinant M-CSF (Peprotech). Monocyte-derived macrophages (MDMs) are preincubated with anti-SIRP α antibodies at the indicated concentrations in 96-well ultra-low attachment TC plates for 30 minutes prior to the addition of fluorescently conjugated microbeads (Spherotech) coated with or without human CD47 (NP_942088; AcroBiosystems) and with or without human IgG1 opsonization to induce Fc-receptor mediated phagocytosis. Following a 1h incubation at 37°C, 5% CO₂, samples are washed, blocked with Human TruStain FcX™ (Biolegend) and stained with fluorescently-conjugated antibody that recognize the external surface of the beads to enable differentiation between beads stuck on the outside of the cells and those that were inside. Cells are washed, fixed, and analyzed by flow cytometry. Phagocytosis is represented as the percentage of cells with internalized beads.

[00303] Internalization of beads (with or without opsonization) by MDMs from donors homozygous for V1 or V2 alleles (Figures 18A-18B and 20A-20B, respectively) or heterozygous for both alleles (Figures 19A-19B) dropped when cells were incubated with CD47-coated beads (white bars); an effect not observed when cells were incubated with beads lacking CD47 (black bars). Pre-treatment of cells with 67 nM of the indicated antibodies restored the cells' ability to phagocytize CD47-coated beads (white bars) with (Figures 18A, 19A, and 20A) and without (Figures 18B, 19B, and 20B) opsonization indicating that the antibodies can block CD47-mediated SIRP α signaling. Further experiments showed that anti-SIRP α antibodies could restore CD47-mediated inhibition of phagocytosis by primary MDMs derived from donors of all genotypes in a dose-dependent manner (Figures 18C, 19C, and 20C).

Example 16. SIRP α antagonist revert inhibition of phagocytosis caused by CD47 in primary human monocyte-derived dendritic cells of any combination of SIRP α alleles

[00304] A flow-based phagocytosis assay was developed to quantify the SIRP α -mediated regulation of phagocytosis induced by CD47. Monocytes enriched from human PBMCs (EasySep™ Monocyte Enrichment Kit, StemCell Technologies) from donors genotyped as homozygous for the SIRP α V1 allele, V2 allele, or heterozygous for both alleles are differentiated into dendritic cells following 5-6 day of culture in ImmunoCult™-ACF Dendritic Cell Medium spiked with ImmunoCult™-ACF Dendritic Cell Differentiation Supplement. Monocyte-derived dendritic cells (MDDCs) are preincubated with anti-SIRP α antibodies at the indicated concentrations in 96-well ultra-low attachment TC plates for 30 minutes prior to the addition of fluorescently conjugated microbeads (Spherotech) coated with or without human CD47 (NP_942088; AcroBiosystems)

and with or without human IgG1 opsonization to induce Fc-receptor mediated phagocytosis. Following a 1h incubation at 37°C, 5% CO₂, samples are washed, blocked with Human TruStain FcX™ (Biolegend) and stained with fluorescently-conjugated antibody that recognize the beads to enable differentiation between beads stuck on the outside of the cells and those that were inside. Cells are washed, fixed, and analyzed by flow cytometry. Phagocytosis is represented as the percentage of cells with internalized beads.

[00305] Internalization of beads (with or without opsonization) by MDDCs from donors homozygous for V1 or V2 alleles (Figures 21A-21B and 23A-23B, respectively) or heterozygous for both alleles (Figures 22A-22B) dropped when cells were incubated with CD47-coated beads (white bars); an effect not observed when cells were incubated with beads lacking CD47 (black bars). Pre-treatment of cells with 67 nM of the indicated antibodies restored the cells' ability to phagocytize CD47-coated beads (white bars) with (Figures 21A, 22A, and 23A) and without (Figures 21B, 22B, and 23B) opsonization indicating that the antibodies can block CD47-mediated SIRPα signaling.

Example 17. Anti-SIRPα antibodies potentiate U-937 phagocytosis of human tumor cells

[00306] The functional activity of select molecules were evaluated in an antibody-dependent cellular phagocytosis assay. Raji cells (Burkitt's lymphoma cell line; ATCC), are labeled with PKH26 Red (Sigma) according to manufacturer's instructions and subsequently opsonized with Rituximab. Raji cells are washed to remove unbound Rituximab. V1- or V2-SIRPα-expressing U-937 cells (50,000) are treated with serially titrated anti-SIRPα mAb in 96-well ultra-low attachment TC plates for 30 minutes prior to the addition of 50,000 Raji target cells (± Rituximab opsonization). Following a 2h incubation in a humidified incubator at 37°C, 5% CO₂, samples are washed, blocked with Human TruStain FcX™ (Biolegend) and stained with PB-conjugated anti-human CD13 (U-937 marker; clone WM15; BD Biosciences) and AF647-conjugated anti-human CD19 (Raji marker; clone HIB19; Biolegend) cocktail. Cells are washed, fixed, and analyzed by flow cytometry. Phagocytosis is represented as the percentage of CD13+ cells that stained negative for CD19 and positive for PKH26.

[00307] The results showed that V1- and V2-SIRPα-expressing U-937 cells were incapable of phagocytizing Raji cells devoid of Rituximab opsonization. Opsonization of Raji cells induced an increase in cellular phagocytosis (about 0.2% to about 6%), which was further enhanced, in a dose-dependent manner, by treatment with anti-SIRPα antibodies.

Example 18. Anti-SIRP α antibodies potentiate monocyte-derived macrophage phagocytosis of human tumor cells

[00308] The functional activity of select molecules were evaluated in an antibody-dependent cellular phagocytosis assay. Raji cells (Burkitt's lymphoma cell line; ATCC), are labeled with PKH26 Red (Sigma) according to manufacturer's instructions and subsequently opsonized with Rituximab. Raji cells are washed to remove unbound Rituximab. Monocytes enriched from human PBMCs (EasySep™ Monocyte Enrichment Kit, StemCell Technologies) from donors genotyped as homozygous for the SIRP α V1 allele, are differentiated into macrophages following 6-7 days in culture in ImmunoCult™-SF macrophage differentiation medium spiked with 50 ng/ml human recombinant M-CSF (Peprotech). Monocyte-derived macrophages (MDMs) are treated with serially titrated anti-SIRP α mAb in 96-well ultra-low attachment U-bottom plates for 30 minutes prior to the addition of 50,000 Raji target cells ($\pm$ Rituximab opsonization). Following a 2h incubation at 37°C, 5% CO₂, samples are washed, blocked with Human TruStain FcX™ (Biolegend) and stained with BV421-conjugated anti-human CD14 (MDM marker; clone M5E2; Biolegend) and AF647-conjugated anti-human CD19 (Raji marker; clone HIB19; Biolegend) cocktail. Cells are washed, fixed, and analyzed by flow cytometry. Phagocytosis is represented as the percentage of CD13+ cells that stained negative for CD19 and positive for PKH26.

[00309] The results showed that MDMs were incapable of phagocytizing Raji cells devoid of opsonization regardless of cell treatment. Opsonization of Raji cells induced an increase in cellular phagocytosis (about 1.3 to about 17%), which was further enhanced by anti-SIRP α antibody treatment.

Example 19. Mixed Lymphocyte Reaction

[00310] Selected molecules are evaluated in a mixed lymphocyte reaction. 96-well round bottom tissue culture treated plates were coated with recombinant human CD47 extracellular domain (ECD)/hFc protein diluted in DPBS and incubated overnight at 4°C. Plates are washed 3X in DPBS/CF. Cryopreserved human monocyte-derived dendritic cells (Astarte Biologics) are thawed, washed and resuspended at 0.1 million/ml in X-VIVO 15 media. MDDCs are then pre-incubated with various anti-SIRP α mAb together with or without anti-PD1 antagonist at a 1:1 volume ratio for 60 minutes prior to transferring MDDC $\pm$ mAb treatment to respective wells of a 96-well round bottom TC plate previously coated with or without human CD47 fusion

protein. Human CD4⁺ T cells (about 100,000), enriched from allogeneic human PBMC donors (EasySep™ Human CD4⁺ T Cell Isolation Kit) are added to respective wells and plates are incubated in a humidified incubator at 37°C, 5% CO₂ for 5 days. Sixteen to eighteen hours prior to harvest, culture wells are pulsed with 1 µCi (10 µl at 100 µCi/ml) of ³[H]thymidine (Moravsek Biochemicals Inc.). Cells are subsequently harvested onto filter mats (Wallac) using the Molecular Devices Micro96 Harvester, dried, and then sealed in sample bags (Perkin Elmer) with 10 ml of BetaPlate Scintillation (Perkin-Elmer, cat#1205-440). ³[H]Thymidine incorporation is measured using a MicroBeta2 2450 Microplate Counter (Perkin Elmer).

[00311] Figures 24A-24B show that platebound human CD47 fusion protein inhibited CD4 T cell proliferation mediated by dendritic cells in Mixed Lymphocyte Reaction, demonstrating that CD47-mediated signaling can be highly immune-suppressive. CD47-Fc mediated immunosuppression in these assays can be reverted to various degrees by anti-SIRPα antibodies (KWAR23, antibody A and E) alone or in combination with anti-PD1 antagonist.

Example 20. Crystallization of anti-SIRPα Fab:SIRPα complexes

[00312] To understand the mechanistic bases for antibody binding to SIRPα, an analysis was performed on the structure of the antibody-SIRPα complexes for Antibody E and Antibody A in complex with domain 1 of human SIRPα-V2 (Figures 25A-25F and Figures 26A-26B).

[00313] In order to obtain complex structures we generated a construct for expression of SIRPαV2 (Amino acids 31-148, comprising an N-terminal GST tag) using the Gateway cloning system. Protein was expressed in Origami B(DE3) cells and purified using standard protocols for GST-tagged proteins. Fractions were analyzed by SDS-PAGE and fractions containing the desired protein were pooled and concentrated to 24 mg/ml in 25 mM HEPES; 100 mM NaCl; 5 % Glycerin; pH 7.0. SIRPαV2 protein was mixed with Fab fragments of Antibody A or E and purified by size-exclusion chromatography in 10 mM Tris; 150 mM NaCl; 1 mM TCEP; pH 7.5. Crystals of SIRPαV2 Fab complexes were obtained by vapor diffusion sitting drop method at a complex concentration of 5 mg/ml (reservoir solution: Antibody A: 20% PEG3350 and 180mM tri-Ammonium citrate and Antibody E: 15% PEG 4000 and 100 mM HEPES, pH 7.0). Data was collected at the Swiss Light Source (SLS) and the structures were solved and modelled at resolutions of 1.4 Å (Antibody A) and 1.6 Å (Antibody E).

[00314] The crystal structure analysis demonstrated that the two antibodies can contact SIRP α on different areas and in different orientations. While both antibodies obstruct the CD47-binding site on SIRP α , Antibody A can contact most of the SIRP α residues that are engaged by CD47 indicating how this antibody can achieve complete CD47 antagonism. The SIRP α residues contacted by these antibodies explained how both alleles of SIRP α can be bound equally without detrimental effects by the variant residues between the alleles. The contact points also explain the selectivity against SIRP γ (Figures 25A-25F and Figures 26A-26B and Tables 51-52).

Table 51: Epitope, contact points on SIRP α -V2 and Antibody A

Residues on SIRPα V2 that contact Antibody A	Residues on the Heavy Chain that contact SIRPα V2	Residues on the Light Chain that contact SIRPα V2
LEU 60	H-SER 31	L-ASP 28
ILE 61	H-TYR 32	L-ASN 30
VAL 63	H-ASP 33	L-ASN 31
GLY 64	H-THR 53	L-TYR 32
PRO 65	H-ARG 97	L-TYR 49
GLN 82	H-GLY 98	L-THR 50
LYS 83	H-GLY 99	L-TYR 91
GLU 84	H-VAL 100	L-VAL 92
THR 97	H-TRP 101	L-TYR 96
LYS 98	H-ASP 102	
ARG 99	H-ASP 103	
GLU 100 (ASN 100 in V1)		
LYS 126		
GLY 127		
SER 128		
PRO 129		
ASP 130		

Table 52: Epitope, contact points on SIRP α -V2 and Antibody E

Residues on SIRP α V2 that contact Antibody E	Residues on the Heavy Chain that contact SIRP α V2	Residues on the Light Chain that contact SIRP α V2
ARG 70	H-ARG 30	L-TYR 31
GLY 71	H-ASN 31	L-SER 32
ALA 72	H-TYR 33	L-ASN 33
GLY 73	H-TYR 52	L-THR 99
PRO 74	H-TYR 53	
ALA 75 (GLY 75 on V1)	H-ASN 54	
ARG 76	H-ARG 56	
GLU 77	H-PHE 58	
ALA 114	H-ALA 100	
ALA 116	H-TYR 101	
GLY 117	H-SER 102	
THR 118	H-GLY 103	
TYR 120	H-ILE 104	
THR 131 (VAL 132 on V1)	H-GLY 105	
GLU 132 (133 on V1)	H-LEU 106	
PHE 133 (134 on V1)		
SER 135 (136 on V1)		
GLU 140 (141 on V1)		

See Figure 26 for SIRP α V1 and SIRP α V2 alignment and numbering.

Example 21. Competition assay with commercial anti-SIRP mAb, clone SE5A5

[00315] Antibodies were assessed for their ability to compete with commercial anti-SIRP mAb, clone SE5A5, binding to V1- and V2-SIRP α -expressing U-937 cells. Cells blocked with Human TruStain FcX™ (Biolegend) are incubated with anti-SIRP α antibodies on ice for 30 minutes prior to the addition of fluorescently conjugated SE5A5 (Biolegend). Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is plotted.

[00316] Figures 27A-27B show that KWAR23 and antibody A blocked the binding of fluorescently conjugated SE5A5 to both V1- and V2-SIRPα expressing U-937 cells. Antibody E blocked SE5A5 binding to V1-SIRPα-expressing U-937 cells, but not V2-SIRPα-expressing cells.

Example 22. Binding of antibodies to full-length cynomolgus SIRPα expressed on CHO cells

[00317] Antibody binding to cells expressing cynomolgus (cyno) SIRPα was evaluated by flow cytometry. CHO-K1 cells expressing full-length cyno SIRPα; derived from protein sequence accession numbers (A) EGM-02252; (B) XP_015313155; or (C) NP_001271679, are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00318] KWAR23 demonstrated dose-dependent binding to all cyno SIRPα-expressing CHO cells lines (H3A9, P3HD10, and HC6) encoding various cynomolgus SIRPα sequences (Figures 28A-28C) with EC₅₀ values ranging between 0.5-2.0 nM (Table 53). Antibodies E and E22 exhibited diverse binding profiles achieving saturable binding curves and EC₅₀ values between 1.1 and 2.5 for clones P3HD10 and HC6 and little to no binding to clone H3A9. While antibodies A and A10 exhibited binding to all 3 clones, binding was strongest to clone H3A9 and up to 5-fold weaker EC₅₀ values for clones P3HD10 and HC6. Titratable binding of antibody A4 was detected on each of the three cell lines but did not achieve saturation at concentrations as high as 200 nM.

Table 53: EC₅₀ of Anti-SIRP α antibodies

Antibody	Cynomolgus SIRP α ; EC ₅₀ ; nM		
	EGM-02252 Clone H3A9	XP_015313155 Clone P3HD10	NP_001271679 Clone HC6
Isotype	NB	NB	NB
KWAR23	0.9	0.5	2
Antibody A	1.9	5	4.5
Antibody E	NS	1.1	2.5
Antibody A10	1.5	7.2	3.7
Antibody A4	NS	NS	NS
Antibody E22	NS	1.3	1.4

^{NB}No binding^{NS}Non-saturable binding**Example 23. Binding of antibodies to full-length cynomolgus SIRP β 1- and SIRP β 1v3 expressed on CHO cells**

[00319] Antibody binding to cells expressing full-length cynomolgus SIRP β 1 (XP_005568598) and SIRP β 1v3 (XP_005568593) is evaluated by flow cytometry. SIRP β 1- or SIRP β 1v3-expressing CHO cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG secondary reagent. Cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00320] KWAR23 demonstrated dose-dependent binding to cyno SIRP β 1- and SIRP β 1v3-expressing CHO cells (Figures 29A and 29B, respectively) with subnanomolar EC₅₀ values (Table 54). Antibodies E and E22 exhibited diverse binding profiles achieving saturable binding curves and subnanomolar EC₅₀ values for SIRP β 1v3-expressing CHO cells (Figure 29B, Table 54), but little to no detectable binding to SIRP β 1-expressing CHO cells (Figure 29A). In contrast, antibodies A and A10 showed dose-dependent binding to SIRP β 1-expressing CHO cells (Figure 29A) with subnanomolar EC₅₀ values (Table 54) and non-saturable binding to SIRP β 1v3-expressing cells (Figure 29B). Dose-dependent binding of antibody A4 was detected on each of two cell lines but did not achieve saturation at concentrations as high as 67 nM.

Table 54: EC₅₀ of Anti-SIRPα antibodies

Antibody	Cynomolgus SIRPβ1; EC ₅₀ , nM	
	SIRPβ1 XP_005568598	SIRPβ1v3 XP-005568593
	Clone PA2	Clone 1HC6
Isotype	NB	NB
KWAR23	0.2	0.4
Antibody A	0.7	NS
Antibody E	NB	0.8
Antibody A10	0.6	NS
Antibody A4	NS	NS
Antibody E22	NB	1

^{NB}No binding^{NS}Non-saturable binding

[00321] The disclosures of each and every patent, patent application, and publication cited herein are hereby incorporated herein by reference in their entirety. While this disclosure has been disclosed with reference to specific embodiments, it is apparent that other embodiments and variations of this disclosure may be devised by others skilled in the art without departing from the true spirit and scope of the invention. The appended claims are intended to be construed to include all such embodiments and equivalent variations.

Example 24. Pharmacokinetics (PK) in cynomolgus monkeys.

[00322] A pharmacokinetic study is conducted in naïve male cynomolgus monkeys (*Macaca fascicularis*) 2 -5 years of age with a body weight range between 2.1 – 6.0 kg. The monkeys are divided into two treatment groups that are administered Antibody A10. Group 1 (n=3) receives 1 mg/kg and Group 2 (n=3) receives 5 mg/kg of the antibody. The antibody is administered intravenously (i.v.) as a 2 mg/ml solution in citrate buffer (50 mM sodium citrate/115 mM sodium chloride, pH 5.0). Blood samples are collected over 6 weeks from a peripheral vein and serum is recovered for analysis.

[00323] Serum samples are analyzed using an ELISA format. Briefly, HuSIRPα-V1 is bound to a NUNC (TM) ELISA plate (Thermo Fisher Scientific). The plates are washed with 0.05% TWEEN (TM) 20 in phosphate buffered saline and blocked with 5% BSA Buffer (Sera Care cat#AP-4510-01), in PBS (Gibco ref 10010-023) prior to incubation with serum samples. The antibody is detected utilizing Novus, pAb anti-goat anti-human IgG HRP, Cat# NB7489 at 1:8000

dilution. PK parameters are determined by non-compartmental analysis using Phoenix WINNONLIN (TM) software (Version 8.2, Certara USA, Inc. Princeton, NJ).

[00324] The PK study results are shown Figure 30 and the PK parameters are summarized in Table 55 below. The antibody demonstrated dose dependent CL between 1 and 5 mg/kg, suggestive of target-mediated drug distribution (TMDD) contributions to overall clearance.

Table 55

Dose (mg/kg)	AUC _{0-inf} (hr*ng/mL)	CL (mL/d/kg)	t _{1/2z} (d)	V _{ss} (mL/kg)	MRT (d)
1.0 IV	101000 (15800)	24.2 (3.54)	1.79 (0.79)	34.7 (6.50)	1.43 (0.157)
5.0 IV	11800000 (4260000)	11.0 (3.79)	3.4 (0.906)	47.4 (4.04)	4.58 (1.24)

Abbreviations: AUC_{0-inf}: area under the curve from dosing to the last measurement and extrapolated to infinity; CL: Clearance; t_{1/2z}: Terminal Half-life; V_{ss}: volume distribution at steady state; MRT: Mean Residence Time

Example 25. Toxicology in cynomolgus monkeys.

[00325] A repeat dose toxicity study is conducted in cynomolgus monkeys with Antibody A10. Parameters evaluated in this study include bioanalysis, toxicokinetic, clinical observation, body weight, food consumption, hematology, blood chemistry, coagulation, urinalysis, immunophenotyping, neurobehavioral examination, electrocardiogram examination, and ophthalmic examination.

[00326] Groups of male and female cynomolgus monkeys (n=3/sex/group) receive the antibody at dose levels of 100 and 250 mg/kg/dose q1w for 4 weeks (5 dose administrations).

[00327] There were no antibody-related changes in body weight, clinical observations, food consumption, ophthalmic examination, neurobehavioral examination, electrocardiogram, immunophenotyping, hematology, coagulation, clinical chemistry, or urinalysis.

[00328] In conclusion, intravenous administration of the antibody at 100 and 250 mg/kg/dose q1w for 4 weeks (5 doses total) was tolerated in monkeys with no antibody-related adverse findings. Under the condition of this study, the no-observed-adverse-effect-level (NOAEL) at steady state (Day 22) was considered to be 250 mg/kg/dose q1w.

Example 26. Binding of antibodies to full-length human V1-SIRP α with various amino acid point mutations expressed on Expi-CHO cells

[00329] To study the structural basis of the observed antibody selectivity, antibody binding to cells engineered to express modified V1-SIRP α protein sequences is tested by flow cytometry. Expi-CHO parental cells are transduced with lentiviruses containing either full-length wild-type human SIRP α V1 allele (NP_542970.1) or full-length human SIRP α V1 with amino acid point mutations (Table 56) and sorted to achieve matching expression levels. These cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG antibodies. Stained cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00330] Antibodies A-E, A4, A10, E22 and KWAR23, but not isotype control, showed dose-dependent binding to Expi-CHO cells expressing full-length wild-type human SIRP α V1 (Figure 31A) with EC₅₀ values ranging between 0.3 to 3.8 nM (Table 57). Antibody binding to cells expressing SIRP α V1 N→E and D→E variants were comparable to binding to SIRP α V1 WT-expressing cells (Figures 31B-31C; Table 57). In contrast, binding of antibodies A-D, A4, and A10 to Expi-CHO cells expressing SIRP α V1 D→N and DD→EN variants was affected, showing abolished, weaker (> EC₅₀), or non-saturable binding (Figures 31D-31E; Table 57). None of the antibodies tested exhibited binding to parental CHO cells (Figure 31F).

Table 56.

Target	Sequence	SEQ ID NO:
α V1 WT	EEELQVIQPDKSVLVAAGETATLRCTATSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRIGNITPADAGT YYCVKFRKGSPPDVEFKSGAGTELSVRAKPSAPVVS GPAAARATP QHTVSFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPVGESVS YSIHSTAKVVLTRDVDHSQVICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTVTENKDGTYNWMSWLLVNVSAHRDDVKLTCQVEHDGQ PAVSKSHDLKVS AHPKEQGSNTAAENTGSNERNIYIVVGVVCTL LVALLMAALYLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTND ITYADLNLPGKKPAPQAAEPNNHTEYASIQTSPPQASEDTLTY ADLDMVHLNRTPKQPAPKPEPSFSEYASVQVPRK	268
α V1 N→E	EEELQVIQPDKSVLVAAGETATLRCTATSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTKR <u>E</u> NNMDFSIRIGNITPADAGT YYCVKFRKGSPPDVEFKSGAGTELSVRAKPSAPVVS GPAAARATP QHTVSFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPVGESVS YSIHSTAKVVLTRDVDHSQVICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTVTENKDGTYNWMSWLLVNVSAHRDDVKLTCQVEHDGQ	269

	PAVSKSHDLKVS AHPKEQGSNTAAENTGSNERNIYIVVGVVCTL LVALLMAALYLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTND ITYADLNLPKGKKPAPQAAEPNNHTEYASIQTSPPQASEDTLTY ADLDMVHLNRTPKQPAPKPEPSFSEYASVQVPRK	
$\alpha V1 D \rightarrow E$	EEELQVIQPDKSVLVAAGETATLRCTATSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTNRNNMDFSIRIGNITPADAGT YYCVKFRKGS PED VEFKSGAGTELSVRAKPSAPVVS GPAARATP QHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPVGESVS YSIHSTAKVVLTR EDVHSQV ICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTVTENKDGTYNWM SWLLVNVSAHRDDVKLTCQVEHDGQ PAVSKSHDLKVS AHPKEQGSNTAAENTGSNERNIYIVVGVVCTL LVALLMAALYLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTND ITYADLNLPKGKKPAPQAAEPNNHTEYASIQTSPPQASEDTLTY ADLDMVHLNRTPKQPAPKPEPSFSEYASVQVPRK	270
$\alpha V1 D \rightarrow N$	EEELQVIQPDKSVLVAAGETATLRCTATSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTNRNNMDFSIRIGNITPADAGT YYCVKFRKGS PDN VEFKSGAGTELSVRAKPSAPVVS GPAARATP QHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPVGESVS YSIHSTAKVVLTR EDVHSQV ICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTVTENKDGTYNWM SWLLVNVSAHRDDVKLTCQVEHDGQ PAVSKSHDLKVS AHPKEQGSNTAAENTGSNERNIYIVVGVVCTL LVALLMAALYLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTND ITYADLNLPKGKKPAPQAAEPNNHTEYASIQTSPPQASEDTLTY ADLDMVHLNRTPKQPAPKPEPSFSEYASVQVPRK	271
$\alpha V1 DD \rightarrow EN$	EEELQVIQPDKSVLVAAGETATLRCTATSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTNRNNMDFSIRIGNITPADAGT YYCVKFRKGS PEN VEFKSGAGTELSVRAKPSAPVVS GPAARATP QHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPVGESVS YSIHSTAKVVLTR EDVHSQV ICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTVTENKDGTYNWM SWLLVNVSAHRDDVKLTCQVEHDGQ PAVSKSHDLKVS AHPKEQGSNTAAENTGSNERNIYIVVGVVCTL LVALLMAALYLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTND ITYADLNLPKGKKPAPQAAEPNNHTEYASIQTSPPQASEDTLTY ADLDMVHLNRTPKQPAPKPEPSFSEYASVQVPRK	272

Table 57.

Antibody	Cell Binding EC ₅₀ (nM)				
	α V1 WT	α V1 N→E	α V1 D→E	α V1 D→N	α V1 DD→EN
Isotype	NB	NB	NB	NB	NB
KWAR23	0.3	0.5	0.2	0.2	0.2
Antibody A	0.4	0.4	0.3	NS	NS
Antibody B	3.8	4.5	3.7	NS	NS
Antibody C	0.3	0.3	0.2	2.7	1.1
Antibody D	0.3	0.3	0.2	9.6	6.6
Antibody E	0.5	0.5	0.4	0.4	0.4
Antibody A4	0.4	0.4	0.3	NS	NS
Antibody A10	0.4	0.4	0.3	NS	NS
Antibody E22	0.8	0.9	0.7	0.8	0.6

NB: No binding

NS: Non-saturable binding at 67 nM

Example 27. Binding of antibodies to full-length human V2-SIRP α with various amino acid point mutations expressed on Expi-CHO cells

[00331] To study the structural basis of the observed antibody selectivity, antibody binding to cells engineered to express modified V2-SIRP α protein sequences is tested by flow cytometry. Expi-CHO parental cells are transduced with lentiviruses containing either full-length wild-type human SIRP α V2 allele (CAA71403.1) or full-length human SIRP α V2 with amino acid point mutations (Table 58) and sorted to achieve matching expression levels. These cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG antibodies. Stained cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00332] Antibodies A-E, A4, A10, E22 and KWAR23, but not isotype control, showed dose-dependent binding to Expi-CHO cells expressing full-length wild-type human SIRP α V2 (Figure 32A) with EC₅₀ values ranging between 0.2 to 3.6 nM (Table 59). Antibody binding to cells expressing SIRP α V2 E→N were comparable to binding to SIRP α V2 WT-expressing cells (Figure 32B; Table 59). In contrast, binding of antibodies A, D, A4 and A10, but not B, C, E, E22, and KWAR23, to Expi-CHO cells expressing SIRP α V2 D→E was affected, showing 3.1 to 7.7 fold greater EC₅₀'s than EC₅₀ values observed for SIRP α V2 WT-expressing cells (Figure 32C; Table 59).

Table 58.

Target	Sequence	SEQ ID NO:
α V2 WT	EEELQVIQPDKSVSVAAGESAILHCTVTSLIPVGPIQWFRGAGPA RELIYNQKEGHFPRVTTVSESTKRENMDFSISISNITPADAGTYY CVKFRKGSPDTEFKSGAGTELSVRAKPSAPVVS GPAARATPQHTV SFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPVGESVSYSIHS TAKVVLTREDVHSQVICEVAHVTLQGDPLRG TANLSETIRVPPTL EVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVSR TETAST VTENKDGTYNWMSWLLVNVSAHRDDVKLT CQVEHDGQPAVSKSHD LKVSAHPKEQGSNTAAENTGSNERNIYIVGVVCTLLVALLMAAL YLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTNDITYADLNL PK GKKPAPQAAEPNNHTEYASIQTSQPASEDTLTYADLDMVHLNRT PKQPAPKPEPSFSEYASVQVPRK	273
α V2 E→N	EEELQVIQPDKSVSVAAGESAILHCTVTSLIPVGPIQWFRGAGPA RELIYNQKEGHFPRVTTVSESTKRENMDFSISISNITPADAGTYY CVKFRKGSPDTEFKSGAGTELSVRAKPSAPVVS GPAARATPQHTV SFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPVGESVSYSIHS TAKVVLTREDVHSQVICEVAHVTLQGDPLRG TANLSETIRVPPTL EVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVSR TETAST VTENKDGTYNWMSWLLVNVSAHRDDVKLT CQVEHDGQPAVSKSHD LKVSAHPKEQGSNTAAENTGSNERNIYIVGVVCTLLVALLMAAL YLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTNDITYADLNL PK GKKPAPQAAEPNNHTEYASIQTSQPASEDTLTYADLDMVHLNRT PKQPAPKPEPSFSEYASVQVPRK	274
α V2 D→E	EEELQVIQPDKSVSVAAGESAILHCTVTSLIPVGPIQWFRGAGPA RELIYNQKEGHFPRVTTVSESTKRENMDFSISISNITPADAGTYY CVKFRKGSPDTEFKSGAGTELSVRAKPSAPVVS GPAARATPQHTV SFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPVGESVSYSIHS TAKVVLTREDVHSQVICEVAHVTLQGDPLRG TANLSETIRVPPTL EVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVSR TETAST VTENKDGTYNWMSWLLVNVSAHRDDVKLT CQVEHDGQPAVSKSHD LKVSAHPKEQGSNTAAENTGSNERNIYIVGVVCTLLVALLMAAL YLVRIRQKKAQGSTSSTRLHEPEKNAREITQDTNDITYADLNL PK GKKPAPQAAEPNNHTEYASIQTSQPASEDTLTYADLDMVHLNRT PKQPAPKPEPSFSEYASVQVPRK	275

Table 59.

Antibody	Cell Binding EC ₅₀ (nM)		
	α V2 WT	α V2 E→N	α V2 D→E
Isotype	NB	NB	NB
KWAR23	0.2	0.2	0.2
Antibody A	0.3	0.3	1.0
Antibody B	3.6	4.1	3.7
Antibody C	0.2	0.2	0.2
Antibody D	0.2	0.2	0.6
Antibody E	0.3	0.3	0.3
Antibody A4	0.2	0.2	1.8
Antibody A10	0.2	0.3	0.7
Antibody E22	0.4	0.4	0.4

NB: No binding

NS: Non-saturable binding at 67 nM

Example 28. Binding of antibodies to full-length human SIRPβ1 with various amino acid point mutations expressed on Expi-CHO cells

[00333] To study the structural basis of the observed antibody selectivity, antibody binding to cells engineered to express modified SIRPβ1 protein sequences is tested by flow cytometry. Expi-CHO parental cells are transduced with lentiviruses containing either full-length wild-type human SIRPβ1 (O00241) or full-length human SIRPβ1 with a single amino acid point mutation (Table 60) and sorted to achieve matching expression levels. These cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG antibodies. Stained cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00334] Antibodies A-E, A4, A10 and KWAR23, but not isotype control or antibody E22, showed dose-dependent binding to full length wild-type human SIRPβ1 expressed on Expi-CHO cells (Figure 33A). Table 61 highlights the binding EC₅₀ calculated for each of the antibodies. While antibodies A, C, D, A4, and A10 exhibited EC₅₀ values ranging between 0.2 to 1.6 nM on wild-type human SIRPβ1-expressing cells, binding of antibodies B and E were non-saturable and no EC₅₀ values were determined. Except for KWAR23, antibody binding was completely lost when a single amino acid substitution (D→H) was introduced within the loop region (Figure 33B).

Table 60.

Target	Sequence	SEQ ID NO:
β1 WT	EDELQVIQPEKSVSVAAGESATLRCAMTSLIPVGPIMWFRGAGA GRELIYNQKEGHFPRVTTVSELTKRNNLDFSISISNITPADAGT YYCVKFRKGSPPDVEFKSGAGTELSVRAPKPSAPVVSGPAVRATP EHTVSFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPAGDSVS YSIHSTARVVLTRGDVHSQVICEIAHITLQGDPLRG TANLSEAI RVPPTLEVTQQPMRAENQANVTCQVSNFYPRGLQLTWLENGNVS RTETASTLIENKDGTYNWSWLLVNTCAHRDDVVLTCQVEHDGQ QAVSKSYALEISAHQKEHGSDITHEAALAPTAPLLVALLLGPKL LLVVGVSIAIYICWKQKA	276
β1 D→H	EDELQVIQPEKSVSVAAGESATLRCAMTSLIPVGPIMWFRGAGA GRELIYNQKEGHFPRVTTVSELTKRNNLDFSISISNITPADAGT YYCVKFRKGSPPD H VEFKSGAGTELSVRAPKPSAPVVSGPAVRATP EHTVSFTCESHGFSPRDITLKWFKNGNELSDFQTNVDPAGDSVS YSIHSTARVVLTRGDVHSQVICEIAHITLQGDPLRG TANLSEAI RVPPTLEVTQQPMRAENQANVTCQVSNFYPRGLQLTWLENGNVS RTETASTLIENKDGTYNWSWLLVNTCAHRDDVVLTCQVEHDGQ QAVSKSYALEISAHQKEHGSDITHEAALAPTAPLLVALLLGPKL LLVVGVSIAIYICWKQKA	277

Table 61.

Antibody	Cell Binding EC ₅₀ (nM)	
	β1 WT	β1 D→H
Isotype	NB	NB
KWAR23	0.2	0.01
Antibody A	1.6	NB
Antibody B	NS	NB
Antibody C	0.2	NB
Antibody D	0.2	NB
Antibody E	NS	NB
Antibody A4	1.5	NB
Antibody A10	1.6	NB
Antibody E22	NB	NB

NB: No binding

NS: Non-saturable binding at 67 nM

Example 29. Binding of antibodies to full-length human SIRPβL with various amino acid point mutations expressed on Expi-CHO cells

[00335] To study the structural basis of the observed antibody selectivity, antibody binding to cells engineered to express modified SIRPβL protein sequences is tested by flow cytometry. Expi-CHO parental cells are transduced with lentiviruses containing either full-length wild-type human SIRPβL (Q5TFQ8) or full-length human SIRPβL with a single amino acid point mutation (Table 62) and sorted to achieve matching expression levels. These cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes,

washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG antibodies. Stained cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00336] Antibodies E, E22 and KWAR23 showed dose-dependent binding to full length human SIRPβL expressed on Expi-CHO cells (Figure 34A) with EC₅₀ values less than 1.2 nM (Table 63). No binding of antibodies A, A4, and A10 could be detected on SIRPβL-expressing cells, while antibodies B, C, and D exhibited non-saturable binding (Figure 34A, Table 63). A single amino acid substitution (H→D) introduced within the loop region maintained, strengthened, or facilitated antibody binding to these cells (Figure 34B, Table 63).

Table 62.

Target	Sequence	SEQ ID NO:
βL WT	EEELQVIQPDKSI SVAAGESATLHCTVTSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTNRNNMDFSIRISNITPADAGT YYCVKFRKGSPDHVEFKSGAGTELSVRAKPSAPVVS GPAARATP QHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPAGDSVS YSIHSTAKVVLTR EDVHSQVICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTLTENKDGTYNWM SWLLVNVS AHRDDVKLTCQVEHDGQ PAVSKSHDLKVS AHPKEQGSNTAPG PALASAAPLLIAFLLGPKV LLVVGVS VIYVWKQKA	278
βL H→D	EEELQVIQPDKSI SVAAGESATLHCTVTSLIPVGPIQWFRGAGP GRELIYNQKEGHFPRVTTVSDLTNRNNMDFSIRISNITPADAGT YYCVKFRKGSPD <u>D</u> VEFKSGAGTELSVRAKPSAPVVS GPAARATP QHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPAGDSVS YSIHSTAKVVLTR EDVHSQVICEVAHVTLQGDPLRG TANLSETI RVPPTLEVTQQPVRAENQVNVTCQVRKFYPQRLQLTWLENGNVS RTETASTLTENKDGTYNWM SWLLVNVS AHRDDVKLTCQVEHDGQ PAVSKSHDLKVS AHPKEQGSNTAPG PALASAAPLLIAFLLGPKV LLVVGVS VIYVWKQKA	279

Table 63.

Antibody	Cell Binding EC ₅₀ (nM)	
	βL WT	βL WT H→D
Isotype	NB	NB
KWAR23	1.0	0.5
Antibody A	NB	0.5
Antibody B	NS	6.7
Antibody C	NS	0.3
Antibody D	NS	0.2
Antibody E	1.0	0.5
Antibody A4	NB	0.5
Antibody A10	NB	0.6

Antibody E22	1.2	0.7
NB: No binding		
NS: Non-saturable binding at 67 nM		

Example 30. Binding of antibodies to full-length human SIRP γ with various amino acid point mutations expressed on Expi-CHO cells

[00337] To study the structural basis of the observed antibody selectivity, antibody binding to cells engineered to express modified SIRP γ protein sequences is tested by flow cytometry. Expi-CHO parental cells are transduced with lentiviruses containing either full-length wild-type human SIRP γ (NP_542970.1) or full-length human SIRP γ with amino acid point mutations (Table 64) and sorted to achieve matching expression levels. These cells are blocked with donkey IgG and incubated on ice with increasing concentrations of antibodies for 60 minutes, washed and stained with AF647-conjugated donkey F(ab')₂ anti-human IgG antibodies. Stained cells are washed, fixed, and analyzed by flow cytometry. Median fluorescent intensity is determined and used as a measure of antibody binding.

[00338] Except KWAR23, none the antibodies tested (Antibodies A-E, A4, A10, E22) showed saturable binding to Expi-CHO cells expressing full-length wild-type human SIRP γ or SIRP γ E $\rightarrow$ D expressing cells (Figures 35A and 35B, respectively). In contrast, antibodies A-D, A4, and A10 showed saturable antibody binding to Expi-CHO cells expressing SIRP γ N $\rightarrow$ D and EN $\rightarrow$ DD variants with all but antibody B with sub nanomolar EC₅₀ values (Figures 35C and 35D, respectively, and Table 65).

Table 64.

Target	Sequence	SEQ ID NO:
γ WT	EEELQMIQPEKLLLVTVGKTATLHCTVTSLLPVGPVLWFRGVGP GRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRISSITPADVGT YYCVKFRKGS PENVEFKSGPGTEMALGAKPSAPVVLGPAARTTP EHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPTGQSV YSIRSTARVVLDPWDVRSQVICEVAHVTLQGDPLRG TANLSEAI RVPPTLEVTQQPMRVGNQVNVTCQVRKFYPQSLQLTWS ENGNVC QRETASTLTENKDGTYNWT SWFLVNISDQRDDVVLTCQVKHDGQ LAVSKRLALEVT VH QKDQSSDATPGPASSLTALLLI AVL LGPIY VPWKQKT	280
γ E $\rightarrow$ D	EEELQMIQPEKLLLVTVGKTATLHCTVTSLLPVGPVLWFRGVGP GRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRISSITPADVGT YYCVKFRKGS <u>P</u> DNVEFKSGPGTEMALGAKPSAPVVLGPAARTTP EHTVSFTCESHGFS PRDITLKWFKNGNELSDFQTNVDPTGQSV	281

	YSIRSTARVVLDPWDVRSQVICEVAHVTLQGDPLRGATANLSEAI RVPPTLEVTQQPMRVGNQVNVTCQVRKFYPQSLQLTWSSENGNVC QRETASTLTENKDGTYNWTWSFLVNIHQDDVVLTCQVKHGDGQ LAVSKRLALEVTVHQDKQSSDATPGPASSLTALLLI AVL LGPIY VPWKQKT	
$\gamma N \rightarrow D$	EEELQMIQPEKLLLVTVGKTATLHCTVTSLLPVGPVLWFRGVGP GRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRISSITPADVGT YYCVKFRKGSPE ~~D~~ VEFKSGPGTEALGAKPSAPVVLGPAARTTP EHTVSFTCESHGFSPRDITLKWFKNELSDFTQTNVDPTGQSV YSIRSTARVVLDPWDVRSQVICEVAHVTLQGDPLRGATANLSEAI RVPPTLEVTQQPMRVGNQVNVTCQVRKFYPQSLQLTWSSENGNVC QRETASTLTENKDGTYNWTWSFLVNIHQDDVVLTCQVKHGDGQ LAVSKRLALEVTVHQDKQSSDATPGPASSLTALLLI AVL LGPIY VPWKQKT	282
$\gamma EN \rightarrow DD$	EEELQMIQPEKLLLVTVGKTATLHCTVTSLLPVGPVLWFRGVGP GRELIYNQKEGHFPRVTTVSDLTKRNNMDFSIRISSITPADVGT YYCVKFRKGSPP ~~DD~~ VEFKSGPGTEALGAKPSAPVVLGPAARTTP EHTVSFTCESHGFSPRDITLKWFKNELSDFTQTNVDPTGQSV YSIRSTARVVLDPWDVRSQVICEVAHVTLQGDPLRGATANLSEAI RVPPTLEVTQQPMRVGNQVNVTCQVRKFYPQSLQLTWSSENGNVC QRETASTLTENKDGTYNWTWSFLVNIHQDDVVLTCQVKHGDGQ LAVSKRLALEVTVHQDKQSSDATPGPASSLTALLLI AVL LGPIY VPWKQKT	283

Table 65.

Antibody	Cell Binding EC ₅₀ (nM)			
	γWT	$\gamma E \rightarrow D$	$\gamma N \rightarrow D$	$\gamma EN \rightarrow DD$
Isotype	NB	NB	NB	NB
KWAR23	0.113	0.3	0.3	0.3
Antibody A	NB	NB	0.4	0.6
Antibody B	NS	NS	4.8	5.3
Antibody C	NS	NS	0.2	0.2
Antibody D	NS	NS	0.3	0.3
Antibody E	NB	NB	NB	NB
Antibody A4	NB	NB	0.5	0.7
Antibody A10	NB	NB	0.5	0.7
Antibody E22	NB	NB	NB	NB

NB: No binding

NS: Non-saturable binding at 67 nM

Example 31. Amino acids 125-132 of SIRP α V1 and 125-131 of SIRP α V2 are determinants for antibody selectivity

[00339] A particular area of the SIRP α epitope bound by Antibody A was identified within the loop region located at amino acids 125-131 of SIRP α V2 (sequence RKGSPDT), corresponding to amino acids 125-132 of SIRP α V1 (sequence RKGSPDDV). We identified this

loop as a determinant for the observed binding properties of Antibody A, including selective binding to SIRP α V1 and SIRP α V2, with no detectable binding to human SIRP γ and human SIRP β L (Table 37). The potential role of this loop in conferring binding selectivity was identified by determination of the crystal structure of Antibody A in complex SIRP α V2 (Example 20), and from the sequence comparison of epitope amino acids in related SIRP isoforms, and was confirmed by binding studies described above (Examples 26-30). Notably, this loop differs in length between SIRP α V1 and SIRP α V2, i.e., 8 amino acids in SIRP α V1 and 7 amino acids in SIRP α V2. Despite this difference in loop length, Antibody A shows similar binding affinity for both SIRP α V1 and SIRP α V2 (see, e.g., Table 37). Figure 36A illustrates how Antibody A binds to this loop from the side, which is believed to allow for specific recognition of the loops with different length in SIRP α V1 and SIRP α V2. We observed that the general configuration of the SIRP α interaction with Antibody A is like the SIRP α interaction with the natural binding partner, CD47, where the loops of SIRP α V1 and SIRP α V2 are approached in a similar orientation (Figure 36B). However, unlike Antibody A, CD47 also binds to SIRP γ , indicating that Antibody A can confer binding selectivity despite this general similarity.

[00340] Even though loops of different length can be recognized by Antibody A, this part of the epitope is nonetheless able to confer the selectivity observed for the SIRP proteins. We identified the sequence motif PDDV in SIRP α V1 as an important difference from SIRP γ and SIRP β L, where the corresponding sequences are PENV and PDHV respectively (Figures 37A-37F).

[00341] To further study the structural basis of the observed selectivity of Antibody A, we tested its ability to bind to cells engineered to expressed modified SIRP protein sequences (see Examples 26-30). The results of these experiments confirmed the importance of the variant residues in this loop to conferring binding selectivity. We observed that exchanging the respective sequence motif in SIRP α V1 (PDDV) to either PDNV or PENV sequence of SIRP γ strongly impaired binding to Antibody A (Figures 31D-31E). Similarly, binding of Antibody A to SIRP β 1 with its PDDV motif was completely abrogated by the exchange to PDHV motif of SIRP β L (Figure 33B). Reciprocally, binding can be restored when exchanging the PENV motif of SIRP γ to either PEDV or PDDV of SIRP α V1 (Figures 35C-35D) or exchanging the PDHV motif of SIRP β L to PDDV of SIRP β 1 (Figure 34B). These results highlight the properties of the epitope, allowing on the one hand for tolerance of two different loop length in the SIRP α proteins, but on the other hand providing high sensitivity to amino acid changes in the binding interface. Together, these properties result in the selectivity pattern described for Antibody A and its variants (e.g., A4 and

A10), namely, strong binding to SIRP α V1, SIRP α V2 and SIRP β 1, complemented by pronounced absence of binding to SIRP γ and SIRP β L.

Example 32. Antibody titer and purity.

[00342] Methods

[00343] Transfection. For transfection, combine heavy chain DNA, light chain DNA, Filler DNA and XBP-1 DNA with Opti-Pro SFM (Thermo Fisher) and sterilize by filtering through 0.2 μ m filter. Prepare cells at 2×10^6 cells/mL or 4×10^6 cells/mL (in BalanCD Transfectory CHO (Irvine Scientific) + 4mM L-Glutamine) for required scale of transfection. Add the calculated volume of TransIT Pro (Mirus) transfection reagent to the prepared Opti-Pro SFM + DNA mix and immediately transfer to the prepared cells. Place the shake flask in incubator at 37°C, 5% CO₂, and 140 rpm or 200rpm shake speed. 24 hours post-transfection, temperature is shifted to 30°C, and Transfectory Supplement and Anti-Clumping Supplement (both Irvine Scientific) are added to the transfected cells. CHO CD Efficient Feed B (Gibco) is added between day 5 or day 7, depending on when glucose levels drop between 2 g/L - 1g/L. The transfected culture is maintained for 7-10 days.

[00344] Harvest by centrifugation and sterile filtration. The clarified cell culture supernatant is sampled for titer by ForteBio/Pall Octet Red 96 instrument with Protein A biosensors. The measured antibody concentration is reported as the expression titer (mg/L).

[00345] Protein A (ProA) Purification: The purification is performed at room temperature (RT) using a GE AKTA Pure system. Each sample is captured from the harvested cell culture fluid (HCCF) by recombinant Protein-A affinity chromatography using MabSelect SuRe resin (GE Healthcare). Chromatography steps and buffer details can be found in Tables 66 and 67, respectively. Protein binds to Protein A at neutral pH at room temperature and is washed with high salt (1M NaCl). Each sample is eluted in an isocratic mode using 30 mM sodium acetate, pH 3.5. Eluted sample is neutralized to pH 5.0 using 1% solution of 3M sodium acetate, pH 9.0. Neutralized protein is sterile filtered with 0.22 μ m filtration system. The concentration is calculated based on the absorbance measurement at 280 nm using a NanoDrop 8000 Spectrophotometer (Thermo Fisher).

Table 66.

Buffer	Composition
Equilibration/Wash 1	DPBS: 8.05mM sodium phosphate, 137mM NaCl, 1.47mM potassium phosphate, 2.6mM potassium chloride, pH 7.4
Wash 2	1M NaCl in DPBS
Wash 3	DPBS: 8.05mM sodium phosphate, 137mM NaCl, 1.47mM potassium phosphate, 2.6mM potassium chloride, pH 7.2
Elution	30mM Sodium Acetate, pH 3.5
Neutralization	3M Sodium Acetate, pH~9
Regeneration	0.25N NaOH
Storage	20 % Ethanol

Table 67.

Step	Buffer/Solution	Volume
Equilibration	DPBS, pH 7.4	≥ 5 CV
Load	Media	varies
Wash 1	DPBS, pH 7.4	≥ 5 CV
Wash 2	DPBS+ 1M NaCl	≥ 5 CV
Wash 3	DPBS, pH 7.4	≥ 5 CV
Elution	30mM sodium acetate, pH 3.5	≥ 5 CV Remark: · OD wavelength 280 nm · Start pool collection when $OD \geq 100$ mAU · End pool collection when $OD \geq 100$ mAU
Neutralization	3M sodium acetate, pH9	1% volume of pool post elution
Pre-regeneration wash	DPBS, pH 7.4	≥ 5 CV Remark: · Ensure the column is equilibrated in DPBS, pH 7.2 by checking pH of the column wash
Regeneration	0.25N NaOH	≥ 5 CV Remark:

		· Ensure the column is washed enough in 0.25N NaOH to clean from sticky contaminants · Don't leave the column in regeneration buffer for ≥ 30 min
Post-regeneration wash	DPBS, pH 7.4	≥ 4 CV Remark: · Ensure the column is equilibrated in DPBS, pH 7.2 by checking pH of the column wash
Storage	20 % Ethanol	≥ 4 CV

[00346] Cation exchange chromatography (CEX). Cation exchange chromatography is performed at room temperature. The buffers and process conditions are summarized in Tables 68 and 69. The Protein A affinity chromatography purified sample is polished using a Poros 50 XS column (Thermo Fisher) using an AKTA Avant chromatography system (Cytiva). The antibody is bound to the column which is pre-equilibrated with 5CV (column volumes) of 50mM sodium acetate, pH5.0, washed with 5CV of the same buffer. The antibody is then eluted using gradient of 0 to 0.5M NaCl in 20 CV. Eluted sample is adjusted to the final ionic strength of ~100 mM NaCl. Protein is sterile filtered into a 50mL falcon tube by 0.22 μ m Steriflip (Millipore) using vacuum filtration system. The antibody concentration is calculated based on the absorbance measured as 280 nm using the NanoDrop 8000 (Thermo Fisher). Purified antibody is stored at 4° C.

Table 68.

Buffer	Composition
Equilibration/Wash	50mM Sodium Acetate, pH 5.0
Elution	50mM Sodium Acetate, 1M NaCl, pH 5.0 Gradient elution in 20CV of 50% B
Regeneration	0.25N NaOH
Storage	20% Ethanol

Table 69.

Step	Buffer	Volume
Equilibration	50mM sodium acetate, pH 5.0	5 CV
Load	50mM sodium acetate, pH 5.0	varies
Wash	50mM sodium acetate, pH 5.0	5 CV
Elutions	50mM Sodium Acetate and 1M NaCl, pH 5.0	20 CV Remark: · OD wavelength 280 nm · Start pool collection when $OD \geq 100$ mAU End pool collection when $OD \leq 100$ mAU
Regeneration	0.25N NaOH	≥ 5 CV
Post-regeneration wash	DPBS, pH 7.4	≥ 5 CV
Storage	20 % Ethanol	≥ 5 CV

[00347] Analytical Size Exclusion Chromatography. Analytical Size Exclusion Chromatography (aSEC) is performed using a Acquity UPLC (Waters, Milford, MA) system using a Protein BEH SEC column 200Å, 1.7µm, 4.6x150mm (Waters part#186005225). Running conditions are as follows: Mobile phase: 50mM Sodium Phosphate, 200mM Arginine and 0.05% Sodium Azide; Flow rate: 0.5ml/min; Runtime: 5 minutes; Sample loading amount: 10µg; Peak detection: A280nm. The results are analyzed to determine the percentage of the antibody present as a monomer, low molecular weight (LMW), and high molecular weight (HMW).

[00348] Low pH hold is performed by adjusting the pH to 3.5 with 1M acetic acid, pH 2.45. After 90 minutes incubation at room temperature, the sample is neutralized to pH 5.0 by 1M Tris HCl, pH 9.0. Final concentration is determined using a NanoDrop 8000 (Thermo Fisher) by measuring absorbance at 280nm. Size purity of the samples is characterized by analytical size exclusion chromatography.

[00349] Results

[00350] The above procedures are used to express and purify antibodies of the present disclosure. Tables 70 and 71 show the titer and size purity results from two different trials for the antibodies as indicated. Table 70 shows the expression titer and results of Protein A purification

and cation exchange chromatography. Repeated rows for a given antibody indicate multiple replicates. Table 71 shows the expression titer and results of Protein A purification. Table 72 shows size purity results for the indicated antibodies following pH 3.5 treatment (2 replicates) compared to the same antibody held at pH 5.0.

Table 70.

Name	Titer (mg/L)	ProA load amount (mg)	Recovery from ProA (%)	Monomer following ProA (%)	CEX Load Amount	Recovery from CEX (%)	Monomer following CEX (%)
A	334	334	82.20%	97.03	264.54	78.40%	100
B	206	206	70.78%	95.66	145.81	60.69%	99.2
C	277	277	82.46%	97.76	228.41	76.18%	99.89
D	227	227	80.61%	98.26	182.98	81.26%	99.95
E	136	136	68.71%	98.05	93.45	74.31%	99.98
A10	231	462	101%	97.32	453	84.41%	99.88
A10	287	287	102%	97.15	292	80.61%	99.91
A10	246.5	247	101.00%	96.02	235	79.40%	99.64
A4	137	274	105.60%	99.44	274	85.19%	99.9
E22	77	385	189.10%	95.99	694	79.24%	99.49

Table 71.

Antibody	Expression Titer (mg/L)	Purified Concentration (mg/mL) following Protein A purification	% Monomer (main peak) following Protein A purification
A	286	3.68	98.57
A1	215.1	3.61	98.62
A2	199.4	3.57	99.11
A3	222.6	3.64	98.78
A4	230.5	3.67	98.69
A5	254.7	3.72	98.13
A6	226.5	3.62	98.59
A7	256.2	3.58	98.37
A8	249.8	3.59	98.58
A9	251.1	3.68	98.03
A10	256.3	3.67	97.54
A12	271.6	3.68	98.57

A13	259.2	3.67	98.39
A14	261.4	3.72	98.48
A15	249.2	3.63	97.56
A16	281.3	3.7	98.64
E	146	3.19	99.09
E1	216.1	3.24	87.86
E2	157.6	3.37	97.12
E3	237.7	3.27	88.32
E4	169.8	3.31	97.58
E5	240.6	3.26	88.92
E6	193.4	3.37	98.51
E7	209.2	3.44	86.42
E8	92.8	3.12	98.9
E9	209.7	3.39	88.79
E10	162.9	3.28	100
E11	58.6	1.95	100
E12	235	3.43	88.83
E13	204.6	3.3	89.09
E14	227.5	3.41	92.98
E15	118	3.21	99.14
E16	245.2	3.35	88.1
E17	84.9	2.93	92.69
E18	183.8	3.35	97.63
E19	230.3	3.39	87.89
E20	208	3.49	89.2
E21	237.5	3.46	91.24
E22	78.5	2.89	97.27

Table 72.

Antibody	Monomer%			Avg. Change in Monomer%	HMW%		
	pH 5.0	pH 3.5 (trial #1)	pH 3.5 (trial #2)		pH 5.0	pH 3.5 (trial #1)	pH 3.5 (trial #2)
A10	97.49	97.37	97.40	0.10	2.52	2.62	2.60

A4	99.29	99.13	98.40	0.53	0.71	0.87	1.60
E22	96.64	96.33	96.98	-0.02	3.36	3.66	3.03

Example 33. Antibody Binding Kinetics

[00351] Methods

[00352] The binding kinetics of anti-SIRP α antibodies to various SIRP α analytes are determined by surface plasmon resonance (SPR) using a Biacore 8K+ (Cytiva). 1x HBS-EP+ (Cytiva) is used as running buffer and for all the dilutions unless stated otherwise. The Series S CM5 sensorchip is activated with a 1:1 mixture of 0.1 M NHS (N-hydroxysuccinimide) and 0.4 M EDC (1-Ethyl-3-(3-dimethylaminopropyl) carbodiimide hydrochloride) for 420 seconds at a flow rate of 10 μ l/min and immobilized with Protein A/G (Thermo Scientific, 10 μ g/ml in 10 mM acetate pH 4.5) for 420 seconds at a flowrate of 10 μ l/min resulting in approximately 3000 RU of Protein A/G on the surface. The sensorchip is then deactivated with 1 M ethanolamine HCl pH 8.5 for 420 seconds at a flowrate of 10 μ l/min.

[00353] Antibodies (approximately 1 μ g/ml) are captured on the Protein A/G surface for 60 seconds at a flowrate of 10 μ l/min resulting in capture levels of approximately 240 RU. Dilutions of each SIRP protein (Extracellular Domain + His-Tag, with the concentrations as indicated) are injected as analytes over the captured antibody for 300 seconds of association at a flowrate of 30 μ l/min, followed by a dissociation for 600 seconds. The surfaces are regenerated with a 30 second injection of 0.85% phosphoric acid at 30 μ l/min. SPR sensorgrams are fit globally to 1:1 Langmuir binding to provide on-rate (k_a), off-rate (k_d), and dissociation constant (K_D) values or fit globally to a steady state affinity to provide K_D in the Biacore Insight Evaluation software.

[00354] Results

[00355] The above procedures are used to measure binding properties of antibodies of the present disclosure. For each analyte used, the highest concentration was as indicated in Example 3 (see Table 36). On-rate, off-rate, and dissociation constant of each indicated antibody for each analyte measured are shown in Tables 73-74.

Table 73.

Antibody	huSIRP α V1			huSIRP α V2		
	k_a (1/Ms)	k_d (1/s)	K_D (nM)	k_a (1/Ms)	k_d (1/s)	K_D (nM)
A	4.97E+05	3.84E-03	7.72	8.07E+05	7.28E-03	9.03
E	4.79E+05	2.03E-04	0.43	5.47E+05	1.81E-04	0.33
A1	7.27E+05	6.25E-03	8.60	8.69E+05	1.62E-02	18.6
A2	7.29E+05	6.85E-03	9.39	8.83E+05	1.84E-02	20.9
A3	7.30E+05	6.61E-03	9.06	9.07E+05	1.78E-02	19.6
A4	7.29E+05	6.17E-03	8.46	8.92E+05	1.63E-02	18.3
A5	6.84E+05	5.98E-03	8.75	9.09E+05	1.62E-02	17.8
A6	7.66E+05	6.34E-03	8.28	9.47E+05	1.71E-02	18.1
A7	7.16E+05	6.72E-03	9.38	9.33E+05	1.90E-02	20.4
A8	7.08E+05	6.04E-03	8.53	8.59E+05	1.62E-02	18.8
A9	4.95E+05	3.63E-03	7.34	7.91E+05	6.52E-03	8.25
A10	5.98E+05	4.02E-03	6.75	6.93E+05	6.98E-03	10.6
A11	5.86E+05	4.00E-03	6.87	6.80E+05	7.09E-03	10.9
A12	4.82E+05	3.78E-03	7.83	7.69E+05	6.89E-03	8.96
A13	4.84E+05	3.52E-03	7.28	7.60E+05	6.35E-03	8.36
A14	4.88E+05	3.97E-03	8.13	8.16E+05	6.48E-03	7.95
A15	5.26E+05	3.71E-03	7.06	8.48E+05	6.71E-03	7.91
A16	4.72E+05	3.77E-03	7.98	7.66E+05	6.30E-03	8.22
E1	4.14E+05	2.99E-04	0.72	5.23E+05	3.30E-04	0.63
E2	4.59E+05	4.19E-04	0.91	5.35E+05	3.67E-04	0.69
E3	3.85E+05	3.71E-04	0.96	4.72E+05	3.66E-04	0.78
E4	4.38E+05	4.33E-04	0.99	5.11E+05	4.21E-04	0.82
E5	3.31E+05	3.36E-04	1.02	4.06E+05	3.55E-04	0.87
E6	3.74E+05	3.86E-04	1.03	4.29E+05	3.75E-04	0.88
E7	3.78E+05	4.53E-04	1.20	4.68E+05	5.52E-04	1.18
E8	3.22E+05	3.90E-04	1.21	4.35E+05	2.36E-04	0.54
E9	5.18E+05	6.31E-04	1.22	5.83E+05	5.48E-04	0.94
E10	4.22E+05	5.18E-04	1.23	4.98E+05	5.96E-04	1.20
E11	3.52E+05	4.98E-04	1.41	4.53E+05	2.58E-04	0.57
E12	4.90E+05	8.19E-04	1.67	5.62E+05	7.09E-04	1.26
E13	3.94E+05	6.71E-04	1.70	4.90E+05	3.38E-04	0.69
E14	4.03E+05	7.43E-04	1.84	4.53E+05	6.15E-04	1.36
E15	4.21E+05	8.02E-04	1.91	5.01E+05	3.69E-04	0.74
E16	4.90E+05	1.06E-03	2.15	5.47E+05	1.05E-03	1.93
E17	4.05E+05	9.55E-04	2.36	5.08E+05	4.05E-04	0.80
E18	4.10E+05	1.01E-03	2.46	4.93E+05	4.43E-04	0.90
E19	3.34E+05	8.43E-04	2.52	4.11E+05	3.99E-04	0.97
E20	4.80E+05	1.64E-03	3.42	5.63E+05	6.27E-04	1.11
E21	4.76E+05	2.12E-03	4.46	5.49E+05	7.61E-04	1.39

Table 74.

Antibody	cynoSIRP α (NP 001271679)			cynoSIRP α (EGM 02252)			cynoSIRP α (XP 015313155)		
	k_a (1/Ms)	k_d (1/s)	K_D (nM)	k_a (1/Ms)	k_d (1/s)	K_D (nM)	k_a (1/Ms)	k_d (1/s)	K_D (nM)
A	8.73E+05	1.38E-01	158	n/a	n/a	258*	n/a	n/a	165*
E	2.35E+05	1.91E-03	8.14	NB			2.41E+05	1.70E-03	7.07
A1	n/a	n/a	780*	n/a	n/a	523*	n/a	n/a	427*
A2	n/a	n/a	887*	n/a	n/a	556*	n/a	n/a	469*
A3	n/a	n/a	857*	n/a	n/a	560*	n/a	n/a	480*
A4	n/a	n/a	669*	n/a	n/a	488*	n/a	n/a	420*
A5	n/a	n/a	665*	n/a	n/a	517*	n/a	n/a	415*
A6	n/a	n/a	684*	n/a	n/a	521*	n/a	n/a	420*
A7	n/a	n/a	741*	n/a	n/a	588*	n/a	n/a	466*
A8	n/a	n/a	639*	n/a	n/a	520*	n/a	n/a	415*
A9	n/a	n/a	346*	n/a	n/a	230*	n/a	n/a	174*
A10	n/a	n/a	388*	n/a	n/a	282*	n/a	n/a	203*
A11	n/a	n/a	410*	n/a	n/a	298*	n/a	n/a	204*
A12	n/a	n/a	384*	n/a	n/a	247*	n/a	n/a	195*
A13	n/a	n/a	330*	n/a	n/a	223*	n/a	n/a	177*
A14	n/a	n/a	316*	n/a	n/a	222*	n/a	n/a	176*
A15	n/a	n/a	317*	n/a	n/a	224*	n/a	n/a	174*
A16	n/a	n/a	306*	n/a	n/a	224*	n/a	n/a	174*
E1	1.83E+05	3.27E-03	17.9	NB			1.90E+05	3.17E-03	16.7
E2	2.54E+05	5.26E-03	20.7	NB			2.60E+05	5.12E-03	19.7
E3	1.68E+05	2.91E-03	17.3	NB			1.70E+05	2.88E-03	16.9
E4	2.22E+05	4.68E-03	21.1	NB			2.28E+05	4.51E-03	19.8
E5	1.23E+05	4.71E-03	38.2	NB			1.21E+05	4.59E-03	37.8
E6	1.89E+05	7.74E-03	41.0	NB			1.75E+05	7.29E-03	41.6
E7	1.58E+05	3.46E-03	21.9	NB			1.59E+05	3.37E-03	21.2
E8	2.04E+05	3.60E-03	17.7	NB			2.07E+05	3.44E-03	16.6
E9	3.21E+05	1.40E-02	43.7	NB			3.24E+05	1.32E-02	40.8
E10	2.16E+05	5.45E-03	25.2	NB			2.22E+05	5.26E-03	23.7
E11	2.67E+05	5.42E-03	20.3	NB			2.75E+05	5.49E-03	20.0
E12	2.79E+05	1.29E-02	46.1	NB			2.97E+05	1.25E-02	42.2
E13	1.97E+05	7.55E-03	38.3	NB			1.90E+05	7.11E-03	37.4
E14	1.90E+05	1.73E-02	90.7	NB			2.00E+05	1.73E-02	86.3
E15	2.43E+05	1.20E-02	49.4	NB			2.45E+05	1.13E-02	46.2
E16	3.13E+05	1.58E-02	50.0	NB			3.00E+05	1.47E-02	48.8
E17	2.82E+05	1.26E-02	44.8	NB			2.92E+05	1.26E-02	43.2
E18	2.24E+05	1.18E-02	52.5	NB			2.26E+05	1.13E-02	50.0
E19	1.56E+05	7.25E-03	46.6	NB			1.54E+05	6.87E-03	44.7
E20	2.51E+05	2.78E-02	111.0	NB			2.51E+05	2.61E-02	104.0
E21	2.25E+05	2.42E-02	107.0	NB			2.28E+05	2.45E-02	108.0

Abbreviations for Tables 73-74. NB: No binding. * indicates steady-state affinity (k_a and k_d values unavailable).

Example 34. Antibody Concentration Effects on Stability

[00356] Antibody A10 is prepared at varying concentrations in 10 mM histidine (pH 6.0). Each prepared sample is assessed for turbidity, viscosity, and for high molecular weight (HMW), antibody monomer, and low molecular weight (LMW) levels by size exclusion chromatography (SEC). SEC is performed initially and after one week at 25°C. Results are shown in Table 75 below.

[00357] Intrinsic fluorescence measurements are used to assess domain unfolding and aggregation formation as a function of temperature. Results are shown in Table 76 below.

Table 75. Antibody properties with varying concentration

Nominal Conc [mg/mL]	Measured Conc [mg/mL] n = 2	Turbidity [FNU] n = 2	Viscosity @ 20°C [mPa*s] n = 2	Initial			1 wk@25°C		
				HMW [%]	Mono [%]	LMW [%]	HMW [%]	Mono [%]	LMW [%]
50	50.12	7.98	1.86	0.2	99.38	0.41	0.3	99.27	0.43
62.5	66.24	7.69	2.6	0.22	99.37	0.41	0.36	99.21	0.43
100	94.35	8.39	4.16	0.27	99.25	0.48	0.46	99.11	0.43
150	144.28	8.37	14.73	0.34	99.22	0.44	0.72	98.81	0.46
200	201.50	9.72	75.66**	0.45	99.09	0.45	1	98.53	0.47
250	229.50	8.79	206.0**	0.5	99.04	0.46	1.13	98.4	0.47

** viscosity measurement n=1. Abbreviations: Conc: Concentration; FNU: Formazin Nephelometric Unit; HMW: High Molecular Weight; Mono.: Monomer; LMW: Low Molecular Weight.

Table 76. Intrinsic fluorescence measurement of antibody aggregation with varying concentration.

Nominal Concentration [mg/mL]	Measured Concentration [mg/mL] n = 2	IF T _{off} [°C] (n = 3)	IF T _{m1} [°C] (n = 3)	IF T _{m2} [°C] (n = 3)	IF T _{agg} [°C] (n = 3)
50	50.12	63.4	69.3	80.0	76.5
62.5	66.24	63.4	69.3	80.3	76.4
100	94.35	63.4	69.2	80.5	75.7
150	144.28	63.6	68.8	80.7	75.4
200	201.50	63.6	68.3	81.0	76.0
250	229.50	63.4	68.1	81.2	74.5

Abbreviations: T_{off}: transition onset point; T_{m1}: first transition mid point; T_{m2}: second transition mid point; T_{agg}: transition onset of aggregation.

[00358] Methods

[00359] *Turbidity.* Turbidity is measured using a turbidity photometer. For each measurement, 130 μL of sample is placed in a single-use borosilicate glass round-bottom cuvette. The irradiation wavelength is 633 nm, and turbidity is measured by right angle light scattering. At least two replicates of each measurement are taken with a permitted deviation of $\leq 2\%$.

[00360] *Viscosity.* Viscosity is measured using the Haake MARS III Rheometer, Thermo Scientific. The measurement parameters are as follows: Cone: C35/1 ($\varnothing$ 35 mm and 1° angle), titanium. Volume=210 μL , undiluted. Data analysis: 100 data points at shear rate 1000 s^{-1} . Temperature: 20°C .

[00361] *Size exclusion chromatography.* Size exclusion chromatography is conducted on UPLC-System e.g. Agilent 1290 Infinity II using the pre-column KrudKatcher Ultra HPLC In-Line Filter $0.5\mu\text{m} \times 0.004\text{ }\mu\text{m}$ ID (Phenomenex, Torrance, CA) and the column Acquity UPLC BEH200SEC $300 \times 4.6\text{ mm}$, (Waters Corp., Milford, MA). The mobile phase is 40 mM NaH_2PO_4 x H_2O , 0.4 M NaClO_4 , pH 6.8 and the flow rate is 0.3 mL/min. The autosampler is maintained at 5°C and the column is at room temperature. The detection is done by UV detector at wavelength of 280 nm. Peakwidth is $> 0,05\text{ min}$ (1 s) (for EmPower) or approx. 10 Hz (other software). The slit is 4 nm. Samples are diluted to 5 mg/mL in the mobile phase. The load amount is 30 μg per injection. The run time is 15 min. Integration is performed automatically with the algorithm Apex Track as default, peak area is approximately within a retention time of 4.5 – 11.5 min, with activation of the “detect shoulders” setting. The peak width is 20 sec and the detection threshold is 14.

[00362] *Intrinsic fluorescence.* Intrinsic fluorescence is measured using the Prometheus NT. 48 nano DSF (NanoTemper Technologies GmbH, Munich, Germany) using a standardized method. The sample volume is 10 μL per capillary, and the sample concentration and formulation is as indicated. Prometheus NT.48 Series nanoDSF Grade Standard Capillaries is used for protein concentrations $> 0.2\text{ mg/mL}$, and Prometheus NT.48 Series nanoDSF Grade High Sensitivity Capillaries for concentrations $\leq 0.2\text{ mg/mL}$. The temperature ramp is $20^\circ\text{C} - 95^\circ\text{C}$ at 1°C/min . The excitation wavelength for intrinsic fluorescence is set to 285 nm and intensity is between 10 % - 30 % (adapted depending on the optimal measuring range). The value of the detection ratio at 350 / 330 nm is plotted as a function against the temperature. The first derivation of this function is used for determination of Tonset and melting temperature, and Tonset is determined by light backscattering.

Example 35. Antibody Stability Testing

[00363] Antibody A10 is formulated at a concentration of 50 mg antibody/mL in 25 mM histidine and pH 6.0, with varying NaCl concentration. Properties of each formulation were assessed by SPR (Table 77) and SEC (Tables 78 and 79) initially and after 4 and 8 weeks held at 40°C.

Table 77. Change in binding activity (by SPR).

NaCl [mM]	Activity [% change from baseline]	
	4 weeks@40°C	8 weeks@40°C
25	84.92	73.35
50	85.63	74.13
75	86.01	74.86
100	86.58	76.08
150	87.64	77.52
200	88.37	78.79
250	89.02	79.95
500	90.71	82.68

Table 78. High molecular weight species levels.

NaCl [mM]	High Molecular Weight [%]	
	4 weeks@40°C	8 weeks@40°C
25	2.57	3.73
50	2.56	3.67
75	2.56	3.57
100	2.36	3.30
150	2.08	2.94
200	2.08	2.79
250	2.37	2.76
500	2.16	2.61

Table 79. Monomer levels.

NaCl [mM]	Monomer [%]	
	4 weeks@40°C	8 weeks@40°C
25	95.26	92.87
50	95.24	92.87
75	95.22	92.94
100	95.39	93.16
150	95.68	93.50
200	95.64	93.61

250	95.35	93.64
500	95.50	93.63

[00364] Methods

[00365] *Surface plasmon resonance.* Surface plasmon resonance spectroscopy is performed using the Biacore T200 (Cytiva, Marlborough, MA). Running- and dilution buffer are HBS-EP+ (Cytiva, Marlborough, MA). The analysis temperature was 25 °C and the data collection rate is 1 Hz using one flow cell. The Sensor Chip CM5 is used with high density Protein A/G immobilization. A predefined standard amine coupling method of the software is applied. The immobilization buffer is acetate at pH 4.5, 30 µg/ml, 420 s, 10 µl/min. Analysis is performed by preparing a calibration curve from 2000 – 62.5 ng/ml with factor 1:2 and additional blank injections. Antibody samples are diluted to 1 µg/ml. Human SIRPα V1 & V2 antigens are diluted to 10 µg/ml. For the build method, one cycle includes: capture of the antibody for 180 s, 10 µl/min; injecting antigen for 180 s, 10 µl/min; and regeneration with 50 mM HCl for 12 s, 50 µl/min. Evaluation of the received sensorgram was performed using the calibration curve to calculate Biacore concentration of samples for Protein A/G and antigen and calculate the binding activity for the antigen binding site.

[00366] *Size exclusion chromatography.* Size exclusion chromatography is performed as described in Example 34, above.

Example 36. Intrinsic biophysical profile analysis

[00367] Methods

[00368] Physicochemical descriptors for the variable regions of selected antibodies are computed according to the methods described in Ahmed *et al.*, *Proc Natl Acad Sci U S A.* 2021. Sep 14;118(37):e2020577118 with a pH value of 7.4 and 137 mM salt. The five descriptors are: surface area buried between VL and VH domains (BSA_VL: VH), ratio of charged to hydrophobic surface patches (RP), ratio of dipole and hydrophobic moments (RM), hydrophobic anisotropy (Avg_HI), and structure-based isoelectric point (pIFv_3D). Each of these five descriptor values was used to compute Z-scores by comparing them with the average and standard deviation (SD) values of the corresponding descriptors for the 79 Fv regions contained in 77 approved antibody-based biotherapeutics (two of the approved biotherapeutics each contain two Fv regions). Each descriptor with a Z-score > 1.96 or < -1.96 contributes a flag for an Fv region. The number of flags was summed for each selected antibody, such that an Fv region can collect up to five flags.

The individual Z-scores for each Fv region are also combined to obtain its Z-distance as described (*id.*).

[00369] Results

[00370] The above procedures are used to evaluate intrinsic biophysical properties of selected antibodies. The variable region sequences of antibodies A-E, A1-A16, and E1-E22 are as disclosed above, and the sequence of antibodies L14, P11, S4, SB6, S7, S10, and S14 are as shown in Tables 80-81. Table 82 shows the values of each of the five descriptors. Table 83 shows the Z-score corresponding to each descriptor value, as well as the Z-distance computed from the five descriptors. Table 84 shows the flag values for each descriptor (0 indicate no flag and 1 indicates a flag, i.e., Z-score > 1.96 or < -1.96) and the total number of flags for each antibody. For a given antibody, higher absolute Z-score values, higher numbers of flags, and higher Z-distance values each indicate greater deviation from the average properties of the 79 Fv regions contained in the 77 approved biotherapeutics used in the analysis.

Table 80. Sequences of Antibody Heavy Chain Variable Regions

Antibody	Heavy Chain Variable Region Sequence	SEQ ID NO:
L14	QVQLVQSGAEVKKPGASVKVSCASGYNFNIYWINWVRQAPGQGL EWIGNIYPSSISTNYNEKFKTRATLTVDKSTSTVYMELSSLRSED TAVYYCARSEGTYYGGRYEGDWFYWGQGTTLTVTVSS	284
P11	EVQLVESGGGLVKPGGSLRLSCAASGFTFSSYMSWVRQTPGKGL EWVATISSGGTYTYYPDSVKGRFTLSRDNAKNSLYLQMNSLRAED TAVYYCASQLTGSEFDYWGQGTTLTVTVSS	286
S4	QSVEESGGRLGTPGTPLTLTCTVSGFSLSSYVMGWFRQAPGKGLE YIGIISSSGSPYYASWVNGRFTISKSTTMDLKMNSPTTEDTATY FCARVGPLGVDYFNIWGPGLTVTVSL	288
SB6	RQLVESGGGLVQPGGSLRLSCTASGFSLSSHGISWVRQAPGKGLE YIGTIGTGIVITYFASWAKGRFTGSKTSSTAYMELSSLRSED TAVY FCARGSAWNPFDPPWGQGTTLTVTVSS	290
S7	RSVEESGGRLVTPGTPLTLTCTVSGFSLSSHGISWVRQAPGKGLE YIGTIGTGIVITYFASWAKGRFTGSKTSTTVDLKITSPTTEDTATY FCARGSAWNPFDPPWGPGTLTVTVSS	292
S10	KVEESGGGLVQPGGSLRLSCAASGFSLSSYVMGWVRQAPGKGLEW VSIISSSGSPYYASWVNGRFTISKDNSEGMVYLQMNSLRAEDTAV YYCARVGPLGVDYFNIWGPQGTTLTVTVSS	294
S14	RQLVESGGGLVQPGGSLRLSCTASGFSLSSHGISWVRQAPGKGLE YIGTIGTGIVITYFASWAKGRFTGSKTSSTAYMELSSLRSED TAVY FCARGSAWNPFDPPWGQGTTLTVTVSS	296

Table 81. Sequences of Antibody Light Chain Variable Regions

Antibody	Light Chain Variable Region Sequence	SEQ ID NO:
L14	DIQMTQSPSSLSASVGDRVTITCRASQDIRNYLNWYQQKPGKAPK LLIYFTSTLHSGVPSRFRSGSGSDFTLTITISLQPEDFATYYCQQ GNTLPWTFGGGTKVEIK	285
P11	DIVITQSPASLAVSLGERATISCRASESVDSYGNSFMHWYQQKPG QPPKLLIYRASNLSEGVDPDRFSGSGSRTDFTLTINPLQAEDVATY YCHQSGDLPWTFGGGTKVEIK	287
S4	DIVMTQTPSSVEAAVGGTVTIKCQAGQSINSYLAWYQQKPGQRPK LLIYYASTLESQVPSRFRSGSGSDYTLTISDLESADAATYYCQS WHYISRSYAFGGGTEVVVK	289
SB6	DIVMTQSPSSLSASVGDRVTITCQASQSVYGNNDLAWYQQKPGQA PKLLIYLASTLATGVPSRFRSGSGSDFTLTITISLQPEDFATYYC LGGGDDEADNTFGQGTKVEIK	291
S7	ALVMTQTPASVSAAVGGTVTTKCQASQSVYGNNDLAWYQHKPGQP PKLLIYLASTLATGVPSRFRSGSGSQFTLTITGVQSDDAATYYC LGGGDDEADNVFGGGTEVVV	293
S10	DIVMTQSPDSLAVSLGERATINCQAGQSINSYLAWYQQKPGQPPK LLIYYASTLESQVDPDRFSGSGSDFTLTITISLQAEDEVAVYYCQS WHYISRSYAFGGGKLEIK	295
S14	DIEMTQSPSSVSASVGDRVTITCQASQSVYGNNDLAWYQQKPGQA PKLLIYLASTLATGVPSRFRSGSGSDFTLTITISLQPEDFATYYC LGGGDDEADNVFGGGTKVEIK	297

Table 82. Intrinsic Biophysical Properties of Antibodies

Antibody	Biophysical Property				
	BSA_LC:HC	RP	RM	Avg_HI	pIFv_3D
A	757.565	2.182	1.052	1.147	7.401
B	880.962	1.78	1.174	1.897	8.426
C	746.141	3.2	1.097	0.822	7.342
D	724.277	2.028	0.794	0.884	7.357
E	814.084	1.605	1.135	0.576	8.001
A1	772.203	1.236	0.567	0.645	7.313
A2	754.856	2.407	0.941	1.007	7.84
A3	749.195	1.6	0.981	0.956	7.65
A4	774.46	1.54	1.03	0.614	7.445
A5	779.811	1.913	1.157	0.578	7.474
A6	811.151	1.946	0.961	0.641	6.771
A7	754.862	2.094	0.949	0.956	7.826
A8	758.408	1.415	1.145	0.571	7.474
A9	722.586	1.472	1.131	0.781	5.415
A10	740.85	1.5	1.048	1.182	7.401
A11	771.402	2.031	1.088	1.083	7.372

A12	763.62	1.882	1.071	1.117	7.386
A13	716.196	1.592	1.303	0.659	6.581
A14	752.533	1.771	1.283	0.629	6.405
A15	731.64	1.228	1.096	0.758	5.783
A16	730.942	1.604	1.298	0.741	6.443
E1	897.824	2	0.972	0.818	7.84
E2	862.05	1.5	1.327	0.624	8.031
E3	880.578	1.786	1.121	0.915	8.397
E4	865.757	1.969	1.241	0.953	8.119
E5	806.757	3	1.123	1.295	8.646
E6	871.934	3.333	1.292	1.147	8.529
E7	858.2	2.152	0.991	0.996	8.368
E8	892.703	2.086	0.745	0.553	7.518
E9	848.808	1.426	1.155	0.659	8.001
E10	849.995	2.194	1.133	1.144	8.397
E11	842.435	1.789	0.796	0.685	7.401
E12	910.73	1.478	1.219	0.883	8.353
E13	827.903	1.744	0.928	0.567	8.148
E14	835.996	2.3	1.314	1.12	8.646
E15	817.36	1.639	1.007	1.031	8.031
E16	874.806	1.795	1.236	0.971	8.397
E17	847.622	1.65	0.94	0.514	7.606
E18	840.334	1.933	1.234	1.14	8.236
E19	871.606	1.634	1.124	0.756	8.294
E20	821.97	1.465	0.903	0.447	7.928
E21	842.136	1.477	1.447	0.956	8.382
E22	886.351	2.793	1.236	0.985	7.987
L14	992.129	1.929	0.654	1.426	8.617
P11	759.501	1.795	1.69	0.685	6.481
S4	893.191	1.829	1.359	1.002	6.756
SB6	895.402	1.784	1.251	1.265	5.846
S7	819.933	2.129	0.765	0.353	5.757
S10	841.792	1.523	0.882	1.133	5.415
S14	871.617	1.822	1.511	1.403	5.313

Table 83. Z-scores and Z-distances Computed from Intrinsic Biophysical Properties of Antibodies

Antibody	Z score					Z-distance
	BSA_LC:HC	RP	RM	Avg_HI	pIFv_3D	
A	-0.813	-0.003	-0.098	0.551	-0.306	1.034
B	0.678	-0.315	0.068	2.503	0.515	2.664
C	-0.952	0.787	-0.038	-0.293	-0.353	1.318

D	-1.216	-0.123	-0.454	-0.131	-0.342	1.354
E	-0.13	-0.451	0.015	-0.933	0.175	1.059
A1	-0.636	-0.737	-0.766	-0.753	-0.377	1.498
A2	-0.846	0.172	-0.252	0.188	0.046	0.92
A3	-0.915	-0.455	-0.197	0.055	-0.107	1.047
A4	-0.609	-0.501	-0.129	-0.833	-0.271	1.186
A5	-0.544	-0.212	0.045	-0.927	-0.248	1.124
A6	-0.166	-0.186	-0.223	-0.764	-0.811	1.164
A7	-0.846	-0.071	-0.241	0.056	0.034	0.885
A8	-0.803	-0.598	0.029	-0.947	-0.248	1.401
A9	-1.236	-0.554	0.01	-0.401	-1.898	2.366
A10	-1.015	-0.532	-0.105	0.643	-0.306	1.354
A11	-0.646	-0.12	-0.05	0.385	-0.33	0.831
A12	-0.74	-0.236	-0.072	0.473	-0.318	0.966
A13	-1.314	-0.461	0.247	-0.718	-0.964	1.855
A14	-0.874	-0.322	0.219	-0.797	-1.104	1.664
A15	-1.127	-0.743	-0.038	-0.46	-1.603	2.146
A16	-1.135	-0.452	0.239	-0.505	-1.074	1.72
E1	0.882	-0.144	-0.209	-0.305	0.046	0.968
E2	0.45	-0.532	0.28	-0.808	0.198	1.121
E3	0.674	-0.311	-0.004	-0.051	0.492	0.891
E4	0.495	-0.168	0.161	0.048	0.269	0.611
E5	-0.219	0.632	-0.001	0.938	0.691	1.343
E6	0.569	0.891	0.232	0.553	0.597	1.354
E7	0.403	-0.027	-0.183	0.159	0.468	0.664
E8	0.82	-0.078	-0.521	-0.994	-0.213	1.409
E9	0.29	-0.59	0.043	-0.716	0.175	0.989
E10	0.304	0.006	0.013	0.544	0.492	0.794
E11	0.213	-0.308	-0.451	-0.649	-0.306	0.927
E12	1.038	-0.549	0.13	-0.134	0.456	1.274
E13	0.037	-0.343	-0.27	-0.958	0.292	1.093
E14	0.135	0.089	0.262	0.483	0.691	0.897
E15	-0.091	-0.425	-0.161	0.25	0.198	0.562
E16	0.604	-0.303	0.155	0.093	0.492	0.855
E17	0.275	-0.416	-0.252	-1.094	-0.142	1.237
E18	0.187	-0.196	0.152	0.534	0.362	0.716
E19	0.565	-0.428	0.001	-0.464	0.409	0.941
E20	-0.035	-0.559	-0.303	-1.268	0.116	1.424
E21	0.209	-0.55	0.444	0.054	0.48	0.881
E22	0.744	0.471	0.154	0.13	0.163	0.918
L14	2.022	-0.2	-0.646	1.278	0.668	2.574
P11	-0.79	-0.303	0.779	-0.649	-1.043	1.683
S4	0.826	-0.277	0.323	0.174	-0.823	1.254

SB6	0.853	-0.312	0.176	0.858	-1.552	2
S7	-0.059	-0.044	-0.494	-1.513	-1.623	2.275
S10	0.205	-0.515	-0.333	0.515	-1.898	2.07
S14	0.565	-0.282	0.533	1.218	-1.979	2.467

Table 84. Flag Values Computed from Intrinsic Biophysical Properties of Antibodies

Antibody	Flag					
	BSA_LC:HC	RP	RM	Avg_HI	pIFv_3D	Total
A	0	0	0	0	0	0
B	0	0	0	1	0	1
C	0	0	0	0	0	0
D	0	0	0	0	0	0
E	0	0	0	0	0	0
A1	0	0	0	0	0	0
A2	0	0	0	0	0	0
A3	0	0	0	0	0	0
A4	0	0	0	0	0	0
A5	0	0	0	0	0	0
A6	0	0	0	0	0	0
A7	0	0	0	0	0	0
A8	0	0	0	0	0	0
A9	0	0	0	0	0	0
A10	0	0	0	0	0	0
A11	0	0	0	0	0	0
A12	0	0	0	0	0	0
A13	0	0	0	0	0	0
A14	0	0	0	0	0	0
A15	0	0	0	0	0	0
A16	0	0	0	0	0	0
E1	0	0	0	0	0	0
E2	0	0	0	0	0	0
E3	0	0	0	0	0	0
E4	0	0	0	0	0	0
E5	0	0	0	0	0	0
E6	0	0	0	0	0	0
E7	0	0	0	0	0	0
E8	0	0	0	0	0	0
E9	0	0	0	0	0	0
E10	0	0	0	0	0	0
E11	0	0	0	0	0	0
E12	0	0	0	0	0	0
E13	0	0	0	0	0	0

E14	0	0	0	0	0	0
E15	0	0	0	0	0	0
E16	0	0	0	0	0	0
E17	0	0	0	0	0	0
E18	0	0	0	0	0	0
E19	0	0	0	0	0	0
E20	0	0	0	0	0	0
E21	0	0	0	0	0	0
E22	0	0	0	0	0	0
L14	1	0	0	0	0	1
P11	0	0	0	0	0	0
S4	0	0	0	0	0	0
SB6	0	0	0	0	0	0
S7	0	0	0	0	0	0
S10	0	0	0	0	0	0
S14	0	0	0	0	1	1

Example 37. Analysis of humanness of antibody sequences

[00371] Methods

[00372] Percentage human identity (“humanness”) values are computed for the variable light and variable heavy sequences of selected antibodies using the methods described in Jones *et al.* MAbs. 2016;8(1):1-9. doi: 10.1080/19420862.2015.1114320. In brief, the humanized sequences are compared to their closest human germline counterparts, and percentage identity is determined for each.

[00373] Results

[00374] The above procedures are used to measure the percentage humanness of the light- and heavy-chain variable regions of selected antibodies. Results are shown in Table 85. Higher % Humanness values indicate greater similarity to human germline antibody sequences.

Table 85. Percentage Humanness Values of Antibodies

Antibody	VH		VL	
	Germline	% Humanness	Germline	% Humanness
A	IGHV3-13*01	96.907	IGKV1-16*02	94.737
B	IGHV1-2*02	98.958	IGKV1-12*01	98.947
C	IGHV3-13*01	96.907	IGKV1-16*02	93.684

D	IGHV3-13*01	94.845	IGKV1-16*02	95.789
E	IGHV4-59*01	86.598	IGKV2-28*01	96
A1	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A2	IGHV3-13*01	95.876	IGKV1-16*02	94.737
A3	IGHV3-13*01	95.876	IGKV1-16*02	94.737
A4	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A5	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A6	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A7	IGHV3-13*01	96.907	IGKV1-16*02	94.737
A8	IGHV3-13*01	95.876	IGKV1-16*02	94.737
A9	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A10	IGHV3-13*01	95.876	IGKV1-16*02	94.737
A11	IGHV3-13*01	95.876	IGKV1-16*02	94.737
A12	IGHV3-13*01	95.876	IGKV1-16*02	94.737
A13	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A14	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A15	IGHV3-13*01	94.845	IGKV1-16*02	94.737
A16	IGHV3-13*01	95.876	IGKV1-16*02	94.737
E1	IGHV4-59*01	87.629	IGKV2-28*01	94
E2	IGHV4-59*01	87.629	IGKV2-28*01	96
E3	IGHV4-59*01	88.66	IGKV2-28*01	94
E4	IGHV4-59*01	88.66	IGKV2-28*01	96
E5	IGHV4-59*01	88.66	IGKV2-28*01	94
E6	IGHV4-59*01	88.66	IGKV2-28*01	96
E7	IGHV4-59*01	89.691	IGKV2-28*01	94
E8	IGHV4-4*08	85.567	IGKV2-28*01	94
E9	IGHV4-59*01	87.629	IGKV2-28*01	96
E10	IGHV4-59*01	89.691	IGKV2-28*01	96
E11	IGHV4-4*08	85.567	IGKV2-28*01	96
E12	IGHV4-59*01	88.66	IGKV2-28*01	96
E13	IGHV4-59*01	87.629	IGKV2-28*01	94
E14	IGHV4-59*01	88.66	IGKV2-28*01	96
E15	IGHV4-59*01	87.629	IGKV2-28*01	96
E16	IGHV4-59*01	89.691	IGKV2-28*01	96
E17	IGHV4-4*08	85.567	IGKV2-28*01	96
E18	IGHV4-59*01	88.66	IGKV2-28*01	96
E19	IGHV4-59*01	88.66	IGKV2-28*01	94
E20	IGHV4-59*01	87.629	IGKV2-28*01	96
E21	IGHV4-59*01	88.66	IGKV2-28*01	96
E22	IGHV4-59*01	88.66	IGKV2-28*01	92
L14	IGHV1-46*02	80.612	IGKV1-39*01	88.421
P11	IGHV3-21*01	88.66	IGKV7-3*01	73.737
S4	IGHV3-64*04	60.215	IGKV1-27*01	70.652

SB6	IGHV3-66*01	67.368	IGKV1-6*01	85.556
S7	IGHV3-53*04	54.348	IGKV1-6*01	65.934
S10	IGHV3-66*01	81.522	IGKV4-1*01	82.653
S14	IGHV3-66*01	67.368	IGKV1-6*01	83.333

What is claimed is:

1. An anti-human SIRP α antibody or an antigen-binding fragment comprising:

a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 34 (H-CDR2); and the amino acid sequence of SEQ ID NO: 35 (H-CDR3), and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 or SEQ ID NO: 37 (L-CDR1); the amino acid sequence of SEQ ID NO: 38 (L-CDR2); and the amino acid sequence of SEQ ID NO: 39 (L-CDR3),

or

b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 1 or SEQ ID NO: 223 (H-CDR1); the amino acid sequence of SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5 or SEQ ID NO: 224 (H-CDR2); and the amino acid sequence of SEQ ID NO: 6 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9 or SEQ ID NO: 225 (L-CDR1); the amino acid sequence of SEQ ID NO: 10, SEQ ID NO: 11 or SEQ ID NO: 226 (L-CDR2); and the amino acid sequence of SEQ ID NO: 12 or SEQ ID NO: 227 (L-CDR3),

or

c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 52 (H-CDR1); the amino acid sequence of SEQ ID NO: 53 (H-CDR2); and the amino acid sequence of SEQ ID NO: 54 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 55 (L-CDR1); the amino acid sequence of SEQ ID NO: 56 (L-CDR2); and the amino acid sequence of SEQ ID NO: 57 (L-CDR3),

or

d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 70 (H-CDR2); and the amino acid sequence of SEQ ID NO: 71 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 (L-CDR1); the amino acid sequence of SEQ ID NO: 72 (L-CDR2); and the amino acid sequence of SEQ ID NO: 39 (L-CDR3),

or

e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 262 (H-CDR1); the amino acid sequence of SEQ ID NO: 87 (H-CDR2); and the amino acid sequence of SEQ ID NO: 88 (H-CDR3); and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 (L-CDR1); the amino acid sequence of SEQ ID NO: 72 (L-CDR2); and the amino acid sequence of SEQ ID NO: 89 (L-CDR3).

2. The anti-human SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises:

a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 34 (H-CDR2); the amino acid sequence of SEQ ID NO: 35 (H-CDR3); and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 233, wherein amino acids X1= D or G and X2 = L or A (L-CDR1); the amino acid sequence of SEQ ID NO: 38, (L-CDR2); the amino acid sequence of SEQ ID NO: 39 (L-CDR3),
or

b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 228(H-CDR1), wherein amino acids X1 = N or D; the amino acid sequence of SEQ ID NO: 229, wherein X1=Y or D, X2=N or T, X3=N or Q, and X4=S or P (H-CDR2); the amino acid sequence of SEQ ID NO: 6 (H-CDR3); and
a light chain variable region comprising the amino acid sequence of SEQ ID NO: 230, wherein X1=K or R, X2= N or T, X3=G or A and X4=N, A or T (L-CDR1); the amino acid sequence of SEQ ID NO: 231, wherein X1=L, Q or G and X2=N or S (L-CDR2); the amino acid sequence of SEQ ID NO: 232, wherein X1=M or G (L-CDR3).

3. The anti-human SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises:

a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 34 (H-CDR2); and the amino acid sequence of SEQ ID NO: 35 (H-CDR3), and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 or SEQ ID NO: 37 (L-CDR1); the amino acid sequence of SEQ ID NO: 38 (L-CDR2); and the amino acid sequence of SEQ ID NO: 39 (L-CDR3).

4. The anti-human SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises:

a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 1 or SEQ ID NO: 223 (H-CDR1); the amino acid sequence of SEQ ID NO: 2, SEQ ID NO: 3, SEQ ID NO: 4, SEQ ID NO: 5 or SEQ ID NO: 224 (H-CDR2); and the amino acid sequence of SEQ ID NO: 6 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 7, SEQ ID NO: 8, SEQ ID NO: 9 or SEQ ID NO: 225 (L-CDR1); the amino acid sequence of SEQ ID NO: 10, SEQ ID NO: 11 or SEQ ID NO: 226 (L-CDR2); and the amino acid sequence of SEQ ID NO: 12 or SEQ ID NO: 227 (L-CDR3).

5. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises:

a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 52 (H-CDR1); the amino acid sequence of SEQ ID NO: 53 (H-CDR2); and the amino acid sequence of SEQ ID NO: 54 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 55 (L-CDR1); the amino acid sequence of SEQ ID NO: 56 (L-CDR2); and the amino acid sequence of SEQ ID NO: 57 (L-CDR3).

6. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises:

a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 33 (H-CDR1); the amino acid sequence of SEQ ID NO: 70 (H-CDR2); and the amino acid sequence of SEQ ID NO: 71 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 (L-CDR1); the amino acid sequence of SEQ ID NO: 72 (L-CDR2); and the amino acid sequence of SEQ ID NO: 39 (L-CDR3).

7. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises:

a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 262 (H-CDR1); the amino acid sequence of SEQ ID NO: 87 (H-CDR2); and the amino acid sequence of SEQ ID NO: 88 (H-CDR3); and

a light chain variable region comprising the amino acid sequence of SEQ ID NO: 36 (L-CDR1); the amino acid sequence of SEQ ID NO: 72 (L-CDR2); and the amino acid sequence of SEQ ID NO: 89 (L-CDR3).

8. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 100, 110, 111, 112, 113, 114, 115, 116, or 117; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 105, 125, or 126.

9. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 104, 118, 119, 120, 121, 122, 123, 124 or 221; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 109, 127, 128, 129, 130 or 222.

10. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof comprises a heavy chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 100, 101, 102, 103, or 104; and a light chain variable region comprising the amino acid sequence of any one of SEQ ID NOS: 105, 106, 107, 108, or 109.

11. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 8, wherein the antibody or antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 100; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 105; or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 110; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or
- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 111; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125; or

- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 112;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125;
or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 113;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125;
or
- f) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 114;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125;
or
- g) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 115;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125;
or
- h) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 116;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125;
or
- i) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 117;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 125;
or
- j) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 111;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126;
or
- k) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 112;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126;
or
- l) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 113;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126;
or
- m) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 114;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126;
or
- n) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 115;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126;
or

- o) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 116;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126;
or
- p) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 117;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 126.

12. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 9, wherein the antibody or antigen-binding fragment thereof comprises: a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 104; and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 109; or

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 118;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 118;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 128;
or
- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 119;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or
- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 119;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129;
or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 120;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or
- f) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 120;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129;
or
- g) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 121;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or
- h) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 122;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or

- i) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 118;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- j) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 121;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129;
or
- k) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 122;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129;
or
- l) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 119;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- m) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 123;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or
- n) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 120;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- o) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 123;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129;
or
- p) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 121;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- q) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 122;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- r) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 124;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 129;
or
- s) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 124;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 127;
or

- t) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 123;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- u) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 124;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 130;
or
- v) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 221;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 222.

13. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 10, wherein the antibody or antigen-binding fragment thereof comprises:

- a) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 100;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 105;
or
- b) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 101;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 106;
or
- c) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 102;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 107; or
- d) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 103;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 108; or
- e) a heavy chain variable region comprising the amino acid sequence of SEQ ID NO: 104;
and a light chain variable region comprising the amino acid sequence of SEQ ID NO: 109.

14. The anti-SIRP α antibody according to claim 8, wherein the antibody comprises a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 131, 138, 139, 140, 141, 142, 143, 144, 145, 146, 148, 149, 150, 151 or 152; and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 174, 181, 182, 183, 184, 185, 186, 187, 188, 189, 190, 191, 192, 193, 194, 195 or 218.

15. The anti-SIRP α antibody according to claim 14, wherein the antibody comprises

- a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 131; and a light chain comprising the amino acid sequence of SEQ ID NO: 174; or
- b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 138; and a light chain comprising the amino acid sequence of SEQ ID NO: 121; or
- c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 139; and a light chain comprising the amino acid sequence of SEQ ID NO: 182; or
- d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 140; and a light chain comprising the amino acid sequence of SEQ ID NO: 183; or
- e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 141; and a light chain comprising the amino acid sequence of SEQ ID NO: 184; or
- f) a heavy chain comprising the amino acid sequence of SEQ ID NO: 142; and a light chain comprising the amino acid sequence of SEQ ID NO: 185; or
- g) a heavy chain comprising the amino acid sequence of SEQ ID NO: 143; and a light chain comprising the amino acid sequence of SEQ ID NO: 186; or
- h) a heavy chain comprising the amino acid sequence of SEQ ID NO: 144; and a light chain comprising the amino acid sequence of SEQ ID NO: 187; or
- i) a heavy chain comprising the amino acid sequence of SEQ ID NO: 145; and a light chain comprising the amino acid sequence of SEQ ID NO: 188; or
- j) a heavy chain comprising the amino acid sequence of SEQ ID NO: 146; and a light chain comprising the amino acid sequence of SEQ ID NO: 189; or
- k) a heavy chain comprising the amino acid sequence of SEQ ID NO: 147; and a light chain comprising the amino acid sequence of SEQ ID NO: 190; or
- l) a heavy chain comprising the amino acid sequence of SEQ ID NO: 148; and a light chain comprising the amino acid sequence of SEQ ID NO: 191; or
- m) a heavy chain comprising the amino acid sequence of SEQ ID NO: 149; and a light chain comprising the amino acid sequence of SEQ ID NO: 192; or
- n) a heavy chain comprising the amino acid sequence of SEQ ID NO: 150; and a light chain comprising the amino acid sequence of SEQ ID NO: 193; or
- o) a heavy chain comprising the amino acid sequence of SEQ ID NO: 151; and a light chain comprising the amino acid sequence of SEQ ID NO: 194; or
- p) a heavy chain comprising the amino acid sequence of SEQ ID NO: 152; and a light chain comprising the amino acid sequence of SEQ ID NO: 195; or
- q) a heavy chain comprising the amino acid sequence of SEQ ID NO: 217; and a light chain comprising the amino acid sequence of SEQ ID NO: 218.

16. The anti-SIRP α antibody according to claim 9, wherein the antibody comprises a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 135, 153, 154, 155, 156, 157, 158, 159, 160, 161, 162, 163, 164, 165, 166, 167, 168, 169, 170, 171, 172, 173 or 219; and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 178, 196, 197, 198, 199, 200, 201, 202, 203, 204, 205, 206, 207, 208, 209, 210, 211, 212, 213, 214, 215, 216 or 220.

17. The anti-SIRP α antibody according to claim 16, wherein the antibody comprises:
- a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 135; and a light chain comprising the amino acid sequence of SEQ ID NO: 178; or
 - b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 153; and a light chain comprising the amino acid sequence of SEQ ID NO: 196; or
 - c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 154; and a light chain comprising the amino acid sequence of SEQ ID NO: 197; or
 - d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 155; and a light chain comprising the amino acid sequence of SEQ ID NO: 198; or
 - e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 156; and a light chain comprising the amino acid sequence of SEQ ID NO: 199; or
 - f) a heavy chain comprising the amino acid sequence of SEQ ID NO: 157; and a light chain comprising the amino acid sequence of SEQ ID NO: 200; or
 - g) a heavy chain comprising the amino acid sequence of SEQ ID NO: 158; and a light chain comprising the amino acid sequence of SEQ ID NO: 201; or
 - h) a heavy chain comprising the amino acid sequence of SEQ ID NO: 159; and a light chain comprising the amino acid sequence of SEQ ID NO: 202; or
 - i) a heavy chain comprising the amino acid sequence of SEQ ID NO: 160; and a light chain comprising the amino acid sequence of SEQ ID NO: 203; or
 - j) a heavy chain comprising the amino acid sequence of SEQ ID NO: 161; and a light chain comprising the amino acid sequence of SEQ ID NO: 204; or
 - k) a heavy chain comprising the amino acid sequence of SEQ ID NO: 162; and a light chain comprising the amino acid sequence of SEQ ID NO: 205; or
 - l) a heavy chain comprising the amino acid sequence of SEQ ID NO: 163; and a light chain comprising the amino acid sequence of SEQ ID NO: 206; or

- m) a heavy chain comprising the amino acid sequence of SEQ ID NO: 164; and a light chain comprising the amino acid sequence of SEQ ID NO: 207; or
- n) a heavy chain comprising the amino acid sequence of SEQ ID NO: 165; and a light chain comprising the amino acid sequence of SEQ ID NO: 208; or
- o) a heavy chain comprising the amino acid sequence of SEQ ID NO: 166; and a light chain comprising the amino acid sequence of SEQ ID NO: 209; or
- p) a heavy chain comprising the amino acid sequence of SEQ ID NO: 167; and a light chain comprising the amino acid sequence of SEQ ID NO: 210; or
- q) a heavy chain comprising the amino acid sequence of SEQ ID NO: 168; and a light chain comprising the amino acid sequence of SEQ ID NO: 211; or
- r) a heavy chain comprising the amino acid sequence of SEQ ID NO: 169; and a light chain comprising the amino acid sequence of SEQ ID NO: 212; or
- s) a heavy chain comprising the amino acid sequence of SEQ ID NO: 170; and a light chain comprising the amino acid sequence of SEQ ID NO: 213; or
- t) a heavy chain comprising the amino acid sequence of SEQ ID NO: 171; and a light chain comprising the amino acid sequence of SEQ ID NO: 214; or
- u) a heavy chain comprising the amino acid sequence of SEQ ID NO: 172; and a light chain comprising the amino acid sequence of SEQ ID NO: 215; or
- v) a heavy chain comprising the amino acid sequence of SEQ ID NO: 173; and a light chain comprising the amino acid sequence of SEQ ID NO: 216; or
- w) a heavy chain comprising the amino acid sequence of SEQ ID NO: 219; and a light chain comprising the amino acid sequence of SEQ ID NO: 220.

18. The anti-SIRP α antibody according to claim 10, wherein the antibody comprises a heavy chain comprising the amino acid sequence of any one of SEQ ID NO: 131, 133, 134, 137 or 135; and a light chain comprising the amino acid sequence of any one of SEQ ID NO: 174, 176, 177, 180, or 178.

19. The anti-SIRP α antibody according to claim 18, wherein the antibody comprises:

- a) a heavy chain comprising the amino acid sequence of SEQ ID NO: 131; and a light chain comprising the amino acid sequence of SEQ ID NO: 174; or
- b) a heavy chain comprising the amino acid sequence of SEQ ID NO: 133; and a light chain comprising the amino acid sequence of SEQ ID NO: 176; or

- c) a heavy chain comprising the amino acid sequence of SEQ ID NO: 134; and a light chain comprising the amino acid sequence of SEQ ID NO: 177; or
- d) a heavy chain comprising the amino acid sequence of SEQ ID NO: 137; and a light chain comprising the amino acid sequence of SEQ ID NO: 180; or
- e) a heavy chain comprising the amino acid sequence of SEQ ID NO: 135; and a light chain comprising the amino acid sequence of SEQ ID NO: 178.

20. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody is a monoclonal antibody.

21. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody is a human antibody.

22. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 1, wherein the antibody or antigen-binding fragment thereof binds to human SIRP α at a $K_D \leq 10$ nM.

23. The anti-SIRP α antibody or antigen-binding fragment thereof according to claim 22, wherein the antibody or antigen-binding fragment thereof binds to human or cynomolgus SIRP α at a $K_D \leq 400$ nM.

24. The anti-SIRP α antibody according to any one of claims 1 to 23 for use as a medicament.

25. A pharmaceutical composition comprising an antibody or antigen-binding fragment according to any one of claims 1 to 23 and a pharmaceutically acceptable carrier.

26. The pharmaceutical composition of claim 25 further comprising one or more additional therapeutic agents.

27. The pharmaceutical composition of claim 26, wherein the one or more additional therapeutic agents are selected from the group consisting of: an anti-PD-1 antibody, an anti-PD-L1 antibody, an anti-EGFR antibody and an anti-HER2 antibody.

28. A method of treating a disease, the method comprising administering the antibody or antigen-binding fragment according to any one of claims 1 to 23 or a pharmaceutical composition of claims 25-27, to a subject in need thereof, wherein the disease is selected from cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis.

29. An anti-SIRP α antibody or antigen-binding fragment according to any one of claims 1 to 23, or a pharmaceutical composition of claims 25-27, for use in treating a disease, wherein the disease is selected from cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis.

30. Use of the anti-SIRP α antibody or antigen-binding fragment according to any one of claims 1 to 23, or a pharmaceutical composition of claims 25-27, in manufacture of a medicament for treating a disease, wherein the disease is selected from cancer, inflammatory disease, autoimmune disease, respiratory disease, infectious disease or fibrosis.

31. The method according to claim 28, the anti-SIRP α antibody or antigen-binding fragment according to claim 29, or the use of the anti-SIRP α antibody or antigen-binding fragment according to claim 30, wherein the disease is cancer.

32. The method according to claim 28, the anti-SIRP α antibody or antigen-binding fragment according to claim 29, or the use of the anti-SIRP α antibody or antigen-binding fragment according to claim 30, wherein said antibody or antigen-binding fragment thereof is administered by a parenteral route, intravenous route or subcutaneous route of administration.

33. An isolated polynucleotide encoding a heavy chain variable region amino and/or a light chain variable region according to any one of claims 8, 9, or 10.

34. An isolated polynucleotide encoding a heavy chain and/or a light chain according to any one of claims 14, 16, or 18.

35. A vector comprising a polynucleotide according to any one of claims 33-34, preferably an expression vector, more preferred a vector comprising the polynucleotide according to the invention in functional association with an expression control sequence.

36. A host cell comprising a polynucleotide according to any one of claims 33-34.
37. The host cell of claim 36, wherein the cell is a mammalian cell.
38. A method for the production of an anti-SIRP α antibody or antigen-binding fragment according to any one of claims 1 to 23, comprising the steps:
- (a) cultivating the host cell under conditions allowing the expression of the anti-SIRP α antibody or antigen-binding fragment; and
 - (b) recovering the anti-SIRP α antibody or antigen-binding fragment.
39. A diagnostic kit or diagnostic method comprising an anti-SIRP α antibody or antigen-binding fragment according to any one of claims 1 to 23, or the use thereof.
40. A human anti-SIRP α antibody or antigen-binding fragment, wherein when bound to SIRP α , the antibody or antigen-binding fragment binds to amino acid residues LEU 60, ILE 61, VAL 63, GLY 64, PRO 65, GLN 82, LYS 83, GLU 84, THR 97, LYS 98, ARG 99, GLU 100, LYS 126, GLY 127, SER 128, PRO 129 and ASP 130. as set forth in SEQ ID NO: 266; or LEU 60, ILE 61, VAL 63, GLY 64, PRO 65, GLN 82, LYS 83, GLU 84, THR 97, LYS 98, ARG 99, ASN 100, LYS 126, GLY 127, SER 128, PRO 129 and ASP 130 as set forth in SEQ ID NO:265, and wherein the antibody blocks binding of any of the antibodies of claims 1-23 to SIRP α .
41. A human anti-SIRP α antibody or antigen-binding fragment, wherein when bound to SIRP α , the antibody or antigen-binding fragment binds to amino acid residues ARG 70, GLY 71, ALA 72, GLY 73, PRO 74, ALA 75, ARG 76, GLU 77, ALA 114, ALA 116, GLY 117, THR 118, TYR 120, THR 131, GLU 132, PHE 133, SER 135 and GLU 140 in SEQ ID NO: 266 or ARG 70, GLY 71, ALA 72, GLY 73, PRO 74, GLY 75, ARG 76, GLU 77, ALA 114, ALA 116, GLY 117, THR 118, TYR 120, VAL 132, GLU 133, PHE 134, SER 136 and GLU 141 as set forth in SEQ ID NO:265, and wherein the antibody blocks binding of any of the antibodies of claims 1-23 to SIRP α .
42. The human anti-SIRP α antibody or antigen-binding fragment of claim 40 or 41, wherein the antibody blocks the binding of anti-SIRP α antibody to CD47 by at least about 80%.
43. A method of manufacturing an antibody comprising the steps of:
- a) culturing a host cell comprising an expression vector comprising an isolated polynucleotide encoding a heavy chain variable region according to any one of claims 8-10 and

comprising a polynucleotide encoding light chain variable region according to any one of claims 8-10 under conditions that allow formation of an antibody; and

b) recovering said antibody.

44. A method of manufacturing an antibody comprising the steps of:

a) culturing a host cell comprising an expression vector comprising an isolated polynucleotide encoding a heavy chain according to claim 14, 16 or 18 and a polynucleotide encoding light chain according to claim 14, 16 or 18 under conditions that allow formation of an antibody; and

b) recovering said antibody.

45. The method of claim 43 or 44, further comprising the step of purifying said antibody.

46. The method of claim 43 or 44, further comprising the step of formulating said antibody into a pharmaceutical composition.