(12) INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(19) World Intellectual Property Organization
International Bureau

(43) International Publication Date
5 January 2017 (05.01.2017)

WIPO | PCT

(10) International Publication Number
WO 2017/004252 A1

(51) **International Patent Classification**:
A61K 38/17 (2006.01) *C07K 14/725* (2006.01)
C07K 14/705 (2006.01) *C12N 5/10* (2006.01)

(21) **International Application Number**:
PCT/US2016/040177

(22) **International Filing Date**:
29 June 2016 (29.06.2016)

(25) **Filing Language**: English

(26) **Publication Language**: English

(30) **Priority Data**:
62/186,865 30 June 2015 (30.06.2015) US

(71) **Applicants**: **THE ARIZONA BOARD OF REGENTS ON BEHALF OF THE UNIVERSITY OF ARIZONA** [US/US]; The University of Arizona, Tech Transfer Arizona, University Services Annex, 4th Floor, P.O. Box 210300A, Tucson, AZ 85721 (US). **JOSLIN DIABETES CENTER, INC.** [US/US]; One Joslin Place, Boston, MA 02215 (US).

(72) **Inventors**: **KUHNS, Michael, S.**; The University of Arizona, 1656 E. Mabel Street, PO Box 245221, Tucson, AZ 85724-5221 (US). **SERWOLD, Thomas**; Joslin Diabetes Center, One Joslin Place, Rm 468A, Boston, MA 02215 (US).

(74) **Agent**: **NGUYEN, Quan**; Nguyen & Tarbet, 4199 Campus Drive, Suite 550, Irvine, CA 92612 (US).

(81) **Designated States** *(unless otherwise indicated, for every kind of national protection available)*: AE, AG, AL, AM, AO, AT, AU, AZ, BA, BB, BG, BH, BN, BR, BW, BY, BZ, CA, CH, CL, CN, CO, CR, CU, CZ, DE, DK, DM, DO, DZ, EC, EE, EG, ES, FI, GB, GD, GE, GH, GM, GT, HN, HR, HU, ID, IL, IN, IR, IS, JP, KE, KG, KN, KP, KR, KZ, LA, LC, LK, LR, LS, LU, LY, MA, MD, ME, MG, MK, MN, MW, MX, MY, MZ, NA, NG, NI, NO, NZ, OM, PA, PE, PG, PH, PL, PT, QA, RO, RS, RU, RW, SA, SC, SD, SE, SG, SK, SL, SM, ST, SV, SY, TH, TJ, TM, TN, TR, TT, TZ, UA, UG, US, UZ, VC, VN, ZA, ZM, ZW.

(84) **Designated States** *(unless otherwise indicated, for every kind of regional protection available)*: ARIPO (BW, GH, GM, KE, LR, LS, MW, MZ, NA, RW, SD, SL, ST, SZ, TZ, UG, ZM, ZW), Eurasian (AM, AZ, BY, KG, KZ, RU, TJ, TM), European (AL, AT, BE, BG, CH, CY, CZ, DE, DK, EE, ES, FI, FR, GB, GR, HR, HU, IE, IS, IT, LT, LU, LV, MC, MK, MT, NL, NO, PL, PT, RO, RS, SE, SI, SK, SM, TR), OAPI (BF, BJ, CF, CG, CI, CM, GA, GN, GQ, GW, KM, ML, MR, NE, SN, TD, TG).

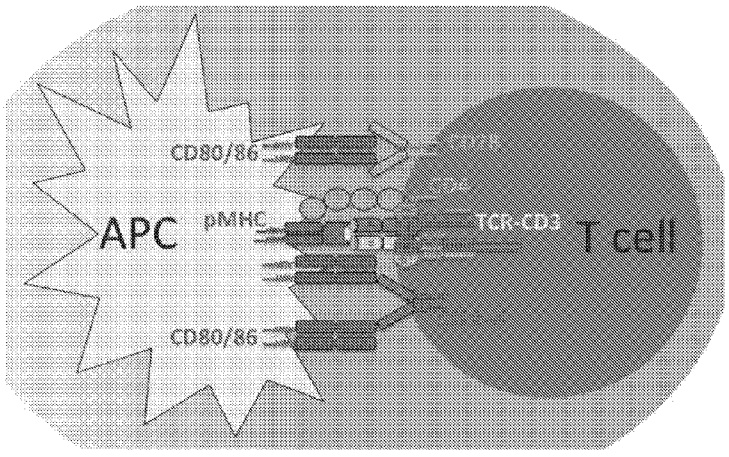
Published:

— *with international search report (Art. 21(3))*

— *with sequence listing part of description (Rule 5.2(a))*

(54) **Title**: REDIRECTED CELLS WITH MHC CHIMERIC RECEPTORS AND METHODS OF USE IN IMMUNOTHERAPY

FIG. 1A



(57) **Abstract**: Chimeric receptors featuring major histocompatibility molecules grafted onto T cell receptor molecules and surrogate co-receptors featuring cell surface receptor ligands fused with signaling molecule domains. The chimeric receptors can be used to redirect cells, altering their specificity. T cells expressing chimeric receptors may bind to ICRs of target T cells for which their chimeric receptors are specific. Surrogate co-receptors may be used to help enhance TCR-CD3 signaling as part of this modular receptor system. The chimeric receptors and surrogate coreceptors may be used to help eliminate autoreactive T cells or program T cells to desired effector functions.

WO 2017/004252 A1

REDIRECTED CELLS WITH MHC CHIMERIC RECEPTORS AND METHODS OF USE IN IMMUNOTHERAPY

CROSS REFERENCE

[0001] This application claims priority to U.S. Provisional Patent Application No. 62/186,865 filed June 30, 2015, the specification(s) of which is/are incorporated herein in their entirety by reference.

REFERENCE TO SEQUENCE LISTING

[0002] Applicant asserts that the information recorded in the form of an Annex C/ST.25 text file submitted under Rule 13*ter*.1(a), entitled UNIA_15_04_PCT_ST25.txt is identical to that forming part of the international application as filed. The content of the sequence listing is incorporated herein by reference in its entirety.

FIELD OF THE INVENTION

[0003] The present invention relates to T cells and T cell receptors, more particularly to redirected T cells with engineered receptors, more particularly to redirected cells expressing a chimeric receptor comprising a major histocompatibility complex (MHC) molecule, including redirected cells further comprising a surrogate coreceptor, e.g., as components of a modular chimeric receptor system.

GOVERNMENT SUPPORT

[0004] This invention was made with government support under Grant No. R01 AI101053 awarded by NIH. The government has certain rights in the invention.

BACKGROUND OF THE INVENTION

[0005] T cells normally recognize and respond to peptide antigens embedded within major histocompatibility complex molecules (pMHCs) of antigen presenting cells (APCs) via their TCR-CD3 complex (see FIG. 1A). This eight-subunit TCR-CD3 complex is composed of the TCR, which is the receptor module that binds the pMHC, and the CD3γε, CD3δε, and CD3ζζ signaling modules that connect the TCR to the intracellular signaling machinery (see FIG. 1B). The intracellular domains of the CD3 subunits contain immunoreceptor tyrosine-based activation motifs (ITAMs) that are phosphorylated by the Src kinases, e.g., Lck, Fyn. CD3γ, CD3δ, and CD3ε each contain one ITAM while CD3ζ contains three ITAMs for a total of ten in a single complex. The TCR-CD3 complex does not appear to have any intrinsic Src kinase activity. In fact,

coreceptors (e.g., CD4, CD8) appear to sequester Lck away from the TCR-CD3 complex until both a coreceptor and a TCR bind a pMHC. The Lck associated with the coreceptor is then brought into close proximity to the CD3 ITAMs to phosphorylate tyrosines within these motifs and initiate signaling.

[0006] Ectopic T cell receptors (TCRs) have been introduced into T cells in an effort to reprogram or alter T cell specificity. However, in some cases, the introduction of ectopic TCRs has been found to lead to cross-pairing events with endogenous TCRs, resulting in novel TCRs with autoimmune specificities. This lead to the use of chimeric antigen receptors (CARs), which are typically designed with (a) an extracellular domain consisting of a single-chain variable fragment (scFv) of a monoclonal antibody directed against a target antigen; (b) a transmembrane domain that does not mediate interactions with other protein subunits; and (c) an intracellular domain consisting of the CD3ζ intracellular signaling domain as well as signaling domains from a variety of other signaling molecules (e.g., CD28, CD27, ICOS, 4-1BB, OX40). Without wishing to limit the present invention to any theory or mechanism, it is believed that CARs do not sufficiently take advantage of the modularity of the existing signaling apparatus, which is optimized to direct T cell activation and effector functions. CARs are likely to be delivering incomplete signals that could have unintended consequences or side effects.

[0007] The present invention features novel chimeric receptors (e.g., "MHCRs") comprising a portion of a MHC molecule (e.g., class I, class II, non-classical MHC) and a portion of the TCR. In some embodiments, the MHCR comprises a portion of an antigen peptide. The present invention also features cells, such as T cells, expressing said MHCRs (cells expressing a MHCR are herein referred to as "redirected cells"). The MHCRs are adapted to recognize and bind to appropriate (specific) TCRs. Redirected cells (e.g., redirected T cells) expressing a MHCR would mimic antigen presenting cells (APCs), the cells that normally express MHC molecules. In some cases, binding of a TCR of a target T cell to the MHCR of the redirected cell may then result in destruction of the target T cell; thus, in this case, the redirected cells may function as "anti-T cell" T cells. The present invention is not limited to redirected cells functioning to destroy a target. For example, in some embodiments, the redirected cell is adapted to help reprogram a target cell, e.g., the redirected cell may deliver instructions to the target cell.

[0008] The present invention also features engineered cells expressing both an MHCR and an SCR. It was surprisingly discovered that engineered cells co-expressing an MHCR and an SCR had enhanced effects (e.g., increased IL-2 expression, see FIG. 5) as compared to engineered cells expressing a MHCR without co-expression of an SCR. Without wishing to limit the present invention to any theory or mechanism, it is believed that the use of an SCR in combination with a MHCR enhances signaling and/or other downstream effects. Without wishing to limit the present invention to any theory or mechanism, it is believed that the combination of the MHCR and SCR may provide a synergistic effect, e.g., effects of the combination of the MHCR and SCR may provide effects greater than those of the MHCR and SCR individually.

[0009] Any feature or combination of features described herein are included within the scope of the present invention provided that the features included in any such combination are not mutually inconsistent as will be apparent from the context, this specification, and the knowledge of one of ordinary skill in the art. Additional advantages and aspects of the present invention are apparent in the following detailed description and claims.

SUMMARY OF THE INVENTION

[0010] The present invention features novel chimeric receptors for engineering redirected cells. For example, the present invention features an engineered cell co-expressing on its surface a chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion (derived from a MHC protein) directly or indirectly fused to a T cell receptor (TCR) portion (derived from a TCR protein); and a surrogate co-receptor (SCR) comprising a cell surface receptor ligand portion directly or indirectly fused to a signaling molecule portion. In some embodiments, the MHCR is adapted to bind to a TCR of a target cell and the SCR is adapted to bind to a cell surface receptor of the target cell. In some embodiments, binding of the MHCR to the TCR of the target cell and binding of the SCR to the cell surface receptor of the target cell (i) initiates a signaling cascade effective for eliminating the target cell or (ii) instructs the target cell to differentiate to a specific effector function. In some embodiments, the cell (e.g., genetically engineered cell) is a T cell (e.g., CD4+, CD8+); however, the present invention is not limited to T cells.

[0011] In some embodiments, the TCR portion comprises a transmembrane domain of

the TCR protein and the MHC portion comprises an extracellular domain of the MHC protein. In some embodiments, the TCR portion comprises at least a portion of a transmembrane domain of the TCR protein and the MHC portion comprises at least a portion of an extracellular domain of the MHC protein. In some embodiments, the TCR portion comprises at least a portion of a transmembrane domain and at least a portion of a cytoplasmic domain of a TCR protein, and the MHC portion comprises at least a portion of an extracellular domain of the MHC protein.

[0012] In some embodiments, the MHC portion of the MHCR is N-terminal to the TCR portion of the MHCR. In some embodiments, the MHC portion is directly fused to the TCR portion. In some embodiments, the MHC portion is indirectly fused to the TCR portion via a linker. In some embodiments, the MHCR further comprises a peptide antigen integrated into the MHC portion, or directly or indirectly fused to the MHC portion. In some embodiments, the peptide antigen is linked to the MHC portion via a linker. In some embodiments, the linker comprises a glycine-rich peptide. In some embodiments, the SCR further comprises a transmembrane domain positioned in between the cell surface receptor ligand portion and the signaling molecule portion. In some embodiments, the MHC protein, the TCR protein, or both the MHC protein and the TCR protein are mammalian proteins (e.g., human, mouse, cat, dog, etc. In some embodiments, the signaling molecule portion has kinase or phosphatase activity. In some embodiments, the signaling molecule portion comprises a Src kinase.

[0013] In some embodiments, the MHC protein comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a fragment thereof, or a combination thereof. In some embodiments, the MHC molecule comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a peptide that is at least 90% identical to HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, or H2-EK beta, a fragment thereof, or a combination thereof. In some embodiments, the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a fragment thereof, or a combination thereof. In some embodiments, the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA,

TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a peptide that is at least 90% identical to TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, or TCC4, a fragment thereof, or a combination thereof. In some embodiments, the cell surface receptor ligand portion of the SCR comprises a CD28 ligand, a CTLA-4 ligand, an ICOS ligand, an OX4O ligand, a PD-1 ligand, or a CD2 ligand. In some embodiments, the CD28 ligand comprises CD80, CD86, or both CD80 and CD86. In some embodiments, the MHCR is adapted to complex with a CD3 subunit. In some embodiments, the engineered cell further co-expresses a second SCR.

[0014] The present invention also features a chimeric receptor (MHCR) as described above. For example, the MHCR may comprise a major histocompatibility complex (MHC) portion derived from a MHC protein directly or indirectly fused to a T cell receptor (TCR) portion derived from a TCR protein, wherein the MHCR is adapted to bind to a TCR of a target cell.

[0015] The present invention also features a method of eliminating a target cell or reprogramming a target cell (the target cell comprising a TCR). In some embodiments, the method comprises introducing a genetically engineered cell that expresses on its surface a chimeric receptor (MHCR) according to the present invention to the target cell, wherein the MHCR is specific for the TCR of the target cell, wherein upon binding of the MHCR to the TCR the genetically engineered cell (a) initiates a signaling cascade that eliminates the target cell, or (b) instructs the target cell to differentiate to a specific effector function. In some embodiments, the method is for immunotherapy. In some embodiments, the target cell is an autoreactive T cell.

[0016] The present invention also features vectors encoding MHCRs of the present invention. The present invention also features vectors encoding SCRs of the present invention.

[0017] Then present invention also features an engineered cell co-expressing on its surface a chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion derived from an extracellular domain of a mammalian MHC protein directly or indirectly linked to a transmembrane domain of a T cell receptor (TCR) portion derived from a mammalian TCR protein, wherein the MHC portion is N-terminal to the TCR portion; and a surrogate coreceptor (SCR) comprising a cell surface receptor

ligand portion indirectly linked to a signaling molecule portion by a transmembrane domain, wherein the signaling molecule portion has kinase or phosphatase activity. The MHCR may be adapted to bind to a TCR of a target cell and the SCR may be adapted to bind to a cell surface receptor of the target cell.

[0018] The present invention also features an engineered T-cell co-expressing on its surface: a chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion derived from an extracellular domain of a mammalian MHC protein directly or indirectly linked to a transmembrane domain of a T cell receptor (TCR) portion derived from a mammalian TCR protein, the MHC portion being N-terminal to the TCR portion, the MHC portion being selected from HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, and H2-EK beta, the TCR portion being selected from TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4; and a surrogate coreceptor (SCR) comprising a cell surface receptor ligand portion indirectly linked to a signaling molecule portion by a transmembrane domain, the signaling molecule portion having kinase or phosphatase activity. The MHCR may be adapted to bind to a TCR of a target cell and the SCR may be adapted to bind to a cell surface receptor of the target cell.

[0019] In some embodiments, the MHC molecule comprises at least a portion of an extracellular domain of a MHC protein. In some embodiments, the TCR molecule comprises at least a portion of a cytoplasmic domain of a TCR protein, at least a portion of a transmembrane domain of a TCR protein, at least a portion of an extracellular domain of a TCR protein, or a combination thereof. In some embodiments, the chimeric receptor is adapted to bind to a TCR. In some embodiments, the chimeric receptor is adapted to complex with at least one CD3 subunit.

[0020] The present invention also features a surrogate co-receptor (SCR) comprising a cell surface receptor ligand portion directly or indirectly fused to a signaling molecule portion via a transmembrane domain, wherein the SCR is adapted to bind to a cell surface receptor of a target cell. In some embodiments, the cell surface receptor ligand portion is indirectly fused to the signaling molecule portion via a linker.

[0021] The present invention also features genetically engineered cells (e.g., redirected

cells) that express on their surfaces a chimeric receptor according to the present invention. In some embodiments, the cell is a T cell (e.g., CD8+ T cell, CD4+ T cell, etc.). In some embodiments, the cell co-expresses one or more SCRs according to the present invention. In some embodiments, the chimeric receptor is complexed with at least one CD3 subunit.

[0022] The present invention also features method of eliminating a target cell or reprogramming a target cell (said target cell comprising a TCR). In some embodiments, the method comprises introducing a genetically engineered cell that expresses on its surface a chimeric receptor to the target cell, wherein the chimeric receptor is specific for the TCR of the target cell. In some embodiments, binding of the chimeric receptor on the genetically engineered cell to the TCR of the target cell initiates a signaling cascade that eliminates the target cell. In some embodiments, binding of the chimeric receptor of the genetically engineered cell to the TCR of the target cell instructs the target cell to differentiate to a specific effector function (e.g. Th1, Th2, Th17, Tfh, Treg or cytotoxic T cell). In some embodiments, the chimeric receptor (e.g., MHCR) is expressed on a Treg and binding of the chimeric receptor to the TCR of a target cell inhibits the target cell's function (e.g., redirect the Treg function against an autoimmune cell). In some embodiments, the genetically engineered cell co-expresses a SCR. In some embodiments, the SCR comprises a cell surface receptor ligand specific for a cell surface receptor on the target cell. In some embodiments, binding of the chimeric receptor to the TCR and binding of the cell surface receptor ligand of the SCR to the cell surface receptor of the target cell initiates a signaling cascade that eliminates the target cell, or instructs the target cell to differentiate to a specific effector function.

[0023] In some embodiments, the method is for immunotherapy. In some embodiments, the genetically engineered cell is surgically introduced to a host (e.g., a mammal). In some embodiments, the target cell is an autoreactive T cell.

[0024] The present invention also features nucleotide sequences encoding the chimeric receptors of the present invention. The present invention also features vectors encoding the chimeric receptors of the present invention. The present invention also features nucleotide sequences encoding the SCRs of the present invention. The present invention also features vectors encoding the SCRs of the present invention.

[0025] Any feature or combination of features described herein are included within the scope of the present invention provided that the features included in any such combination are not mutually inconsistent as will be apparent from the context, this specification, and the knowledge of one of ordinary skill in the art. Additional advantages and aspects of the present invention are apparent in the following detailed description and claims.

BRIEF DESCRIPTION OF THE DRAWINGS

[0026] The features and advantages of the present invention will become apparent from a consideration of the following detailed description presented in connection with the accompanying drawings in which:

[0027] FIG. 1A shows molecules involved in T cell activation. Engagement of the TCR with pMHC (MHC with a peptide antigen) initiates T cell activation.

[0028] FIG. 1B shows the molecular components of the □lpha-beta-TCR-CD3 complex. The TCR transfers pMHC-specific information to the CD3 subunits and inside the T cell. Triangles represent the inner and outer leafs of the cell membrane. Red and blue dots and ovals represent the transmembrane charge interactions that drive subunit assembly of the complexes (from Kuhns et al., 2006, Immunity 24:133-139).

[0029] FIG. 2A shows a redirected T cell expressing a MHCR (pMHCR with peptide antigen) of the present invention. The MHCR in complex with CD3 subunits is bound to a target T cell's TCR.

[0030] FIG. 2B shows non-limiting examples of MHCR configurations (and the schematics are not limiting with respect to N-terminal and C-terminal orientation). TCR refers to the T cell receptor portion; MHC refers to the major histocompatibility portion, antigen refers to the antigen portion, and L refers to a linker. The present invention is not limited to these configurations. For example, in some embodiments the antigen portion is integrated into the MHC portion. In some embodiments, the MHC portion is N-terminal to the TCR portion (see orientation of sequences below).

[0031] FIG. 3A is a schematic view of a chimeric surrogate coreceptor (SCR), e.g., one comprising CD80/CD86-Lck.

[0032] FIG. 3B shows a redirected T cell expressing a MHCR (pMHCR) and two surrogate coreceptors (SCRs). The MHCR, bound to a target T cell's TCR, is complexed with CD3. The SCRs are bound to the target T cell's coreceptors (CD28, CTLA-4). Binding of the SCRs to coreceptors on the target T cell may help initiate CD3 signaling

similar to that seen in normal T cell activation.

[0033] FIG. 4 shows expression of pMHCR-CD3 complexes on T cell hybridomas. 58α⁻β⁻ cells that lack endogenous TCRs were transduced with a pMHCR composed of MCC:I-E^k. The proportional expression (diagonal) of I-E^k and CD3 subunits suggests surface co-dependent expression of the epitopes.

[0034] FIG. 5 shows TCR-specific IL-2 production by pMHCR-CD3 expressing T cell hybridomas. 58α⁻β⁻ cells that lack endogenous TCRs were transduced with a pMHCR composed of MCC:I-E^k as well as a CD80-Lck surrogate coreceptor (SCR). The cells were co-cultured with parental M12 B cells, or M12 cells stably transduced to express the MCC:I-E^k-specific 2B4 TCR alone or with CD28. The increased IL-2 expression in the presence of CD28 indicates that the surrogate coreceptor (SCR) enhances pMHCR-CD3 signaling.

[0035] FIG. 6 shows TCR-specific killing of CD4 T cells by redirected CTLs. Purified CD8 T cells from B10.A mice were activated *in vitro* and transduced with a MCC:I-E^k pMHCR (agonist) or an HB:I-E^k pMHCR (null) as well as a CD80-Lck surrogate coreceptor. The redirected CTLs were then co-cultured at the indicated ratios with naïve *ex vivo* 5c.c7 TCR transgenic CD4 T cells overnight. Killing was evaluated by flow cytometry using count beads relative to the 0:1 samples.

DETAILED DESCRIPTION OF THE INVENTION

Chimeric MHC Receptors (MHCRs)

[0036] The present invention features chimeric receptors (e.g., "MHCRs") comprising at least a MHC portion (e.g., class I, class II, non-classical, a combination thereof, etc.) and a TCR portion (e.g., αβ, γδ TCR, etc.) (see FIG. 2B(i)). For example, the MHCR may comprise a MHC portion and a TCR portion, a MHC and a TCR portion optionally separated by a linker (see FIG. 2B (iii) and (iv)). A linker may be any appropriate linker such as but not limited to a peptide linker. In some embodiments, the MHCR further comprises a peptide antigen (see FIG. 2B (ii)); a MHCR comprising a peptide antigen may herein be referred to as a "pMHCR". Note that MHC portions and/or TCR portions may be from any appropriate species including but not limited to human, monkey, mouse, rat, rabbit, or the like, e.g., any other appropriate mammalian species. The components and configurations of the MHRCs of the present invention are not limited to those shown in FIG. 2B. For example, the MHCR may comprise a TCR portion and a MHC portion; a TCR portion and a MHC portion separated by a linker; a TCR portion

and a MHC portion and an antigen portion; a TCR portion and a MHC portion and an antigen portion, wherein the TCR portion and MHC portion are separated by a linker; a TCR portion and a MHC portion and an antigen portion, wherein the MHC portion and antigen portion are separated by a linker; a TCR portion and a MHC portion and an antigen portion, wherein the TCR and MHC portion are separated by a linker and the MHC portion and the antigen portion are separate by a linker; etc.

[0037] The MHC portion may comprise one or more MHC proteins (e.g., HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB1), one or more fragments thereof, or combinations thereof. For reference, non-limiting MHC sequences (human, mouse) are listed below in Table 1.1 and Table 1.2. Note that MHC genes are highly polymorphic, and thus the present invention is not limited to the sequences in Table 1.1 And Table 1.2. The present invention includes MHC polymorphisms and any other appropriate variant of MHC proteins.

[0038] Table 1.1. Examples of Human MHC Protein Sequences

SEQ ID NO.	Description	Amino Acid Sequence
1	Uniprot P01891 HLA-A gene (MHC I)	MAVMAPRTLV LLLSGALALT QTWAGSHSMR YFYTSVSRPG RGEPRFIAVG YVDDTQFVRF DSDAASQRME PRAPWIEQEG PEYWDRNTRN VKAQSQTDRV DLGTLRGYYN QSEAGSHTIQ MMYGCDVGSD GRFLRGYRQD AYDGKDYIAL KEDLRSWTAA DMAAQTTKHK WEAAHVAEQW RAYLEGTCVE WLRRYLENGK ETLQRTDAPK THMTHHAVSD HEATLRCWAL SFYPAEITLT WQRDGEDQTQ DTELVETRPA GDGTFQKWVA VVVPSGQEQR YTCHVQHEGL PKPLTLRWEP SSQPTIPIVG IIAGLVLFGA VITGAVVAAV MWRRKSSDRK GGSYSQAASS DSAQGSDVSL TACKV
2	Uniprot P18464 HLA-B gene (MHC I)	MRVTAPRTVL LLLWGAVALT ETWAGSHSMR YFYTAMSRPG RGEPRFIAVG YVDDTQFVRF DSDAASPRTE PRAPWIEQEG PEYWDRNTQI FKTNTQTYRE NLRIALRYYN QSEAGSHTWQ

		TMYGCDVGPD GRLLRGHNQY AYDGKDYIAL NEDLSSWTAA DTAAQITQRK WEAAREAEQL RAYLEGLCVE WLRRHLENGK ETLQRADPPK THVTHHPVSD HEATLRCWAL GFYPAEITLT WQRDGEDQTQ DTELVETRPA GDRTFQKWAA VVVPSGEEQR YTCHVQHEGL PKPLTLRWEP SSQSTIPIVG IVAGLAVLAV VVIGAVVATV MCRRKSSGGK GGSYSQAASS DSAQGSDVSL TA
3	Uniprot Q29963 HLA-C gene (MHC I)	MRVMAPRTLI LLLSGALALT ETWACSHSMR YFDTAVSRPG RGEPRFISVG YVDDTQFVRF DSDAASPRGE PRAPWVEQEG PEYWDRETQK YKRQAQADRV NLRKLRGYYN QSEDGSHTLQ WMYGCDLGPD GRLLRGYDQS AYDGKDYIAL NEDLRSWTAA DTAAQITQRK WEAAREAEQW RAYLEGTCVE WLRRYLENGK ETLQRAEHPK THVTHHPVSD HEATLRCWAL GFYPAEITLT WQRDGEDQTQ DTELVETRPA GDGTFQKWAA VVVPSGEEQR YTCHVQHEGL PEPLTLRWEP SSQPTIPIVG IVAGLAVLAV LAVLGAVMAV VMCRRKSSGG KGGSCSQAAS SNSAQGSDES LIACKA
4	Uniprot P20036 HLA DPA1 (MHC II)	MRPEDRMFHI RAVILRALSL AFLLSLRGAG AIKADHVSTY AAFVQTHRPT GEFMFEFDED EMFYVDLDKK ETVWHLEEFG QAFSFEAQGG LANIAILNNN LNTLIQRSNH TQATNDPPEV TVFPKEPVEL GQPNTLICHI DKFFPPVLNV TWLCNGELVT EGVAESLFLP RTDYSFHKFH YLTFVPSAED FYDCRVEHWG LDQPLLKHWE AQEPIQMPET TETVLCALGL VLGLVGIIVG TVLIIKSLRS GHDPRAQGTL
5	Uniprot P04440 HLA DPB1 (MHC II)	MMVLQVSAAP RTVALTALLM VLLTSVVQGR ATPENYLFQG RQECYAFNGT QRFLERYIYN REEFARFDSD VGEFRAVTEL GRPAAEYWNS QKDILEEKRA VPDRMCRHNY ELGGPMTLQR

		RVQPRVNVSP SKKGPLQHHN LLVCHVTDFY PGSIQVRWFL NGQEETAGVV STNLIRNGDW TFQILVMLEM TPQQGDVYTC QVEHTSLDSP VTVEWKAQSD SARSKTLTGA GGFVLGLIIC GVGIFMHRRS KKVQRGSA
6	Uniprot P01909 HLA DQA1 (MHC II)	MILNKALMLG ALALTTVMSP CGGEDIVADH VASYGVNLYQ SYGPSGQYTH EFDGDEQFYV DLGRKETVWC LPVLRQFRFD PQFALTNIAV LKHNLNSLIK RSNSTAATNE VPEVTVFSKS PVTLGQPNIL ICLVDNIFPP VVNITWLSNG HSVTEGVSET SFLSKSDHSF FKISYLTLLP SAEESYDCKV EHWGLDKPLL KHWEPEIPAP MSELTETVVC ALGLSVGLVG IVVGTVFIIR GLRSVGASRH QGPL
7	Uniprot P01920 HLA DQB1 (MHC II)	MSWKKALRIP GGLRAATVTL MLAMLSTPVA EGRDSPEDFV YQFKAMCYFT NGTERVRYVT RYIYNREEYA RFDSDVEVYR AVTPLGPPDA EYWNSQKEVL ERTRAELDTV CRHNYQLELR TTLQRRVEPT VTISPSRTEA LNHHNLLVCS VTDFYPAQIK VRWFRNDQEE TTGVVSTPLI RNGDWTFQIL VMLEMTPQHG DVYTCHVEHP SLQNPITVEW RAQSESAQSK MLSGIGGFVL GLIFLGLGLI IHHRSQKGLL H
8	Uniprot P01903 HLA DRA gene (MHC II)	MAISGVPVLG FFIIAVLMSA QESWAIKEEH VIIQAEFYLN PDQSGEFMFD FDGDEIFHVD MAKKETVWRL EEFGRFASFE AQGALANIAV DKANLEIMTK RSNYTPITNV PPEVTVLTNS PVELREPNVL ICFIDKFTPP VVNVTWLRNG KPVTTGVSET VFLPREDHLF RKFHYLPFLP STEDVYDCRV EHWGLDEPLL KHWEFDAPSP LPETTENVVC ALGLTVGLVG IIIGTIFIIK GVRKSNAAER RGPL
9	Uniprot Q30167 HLA DRB1 gene	MVCLRLPGGS CMAVLTVTLM VLSSPLALAG DTRPRFLEEV KFECHFFNGT ERVRLLERRV

	(MHC II)	HNQEEYARYD SDVGEYRAVT ELGRPDAEYW NSQKDLLERR RAAVDTYCRH NYGVGESFTV QRRVQPKVTV YPSKTQPLQH HNLLVCSVNG FYPGSIEVRW FRNGQEEKTG VVSTGLIQNG DWTFQTLVML ETVPQSGEVY TCQVEHPSVM SPLTVEWRAR SESAQSKMLS GVGGFVLGLL FLGAGLFIYF RNQKGHSGLP PTGFLS

[0039] Table 1.2. Examples of Mouse MHC Protein Sequences

SEQ ID NO.	Description	Amino Acid Sequence
10	Uniprot Q9TQ72 MHC II antigen IE alpha (H2-Aa)	RSRALILGVL ALTTMLSLCG GEDYIEADHV AFYGISVYQS PGDIGQYTFE FDGDELFYVD LDKKETVWML PEFGQLTSFD PQGGLQEIAT GKYNLEILIK DSNFTPAANE APQATVFPKS PVLLGQPNTL ICFVDNIFPP VINITWLRNS KSVTDGVYET SFLVNRDHSF HKLSYLTFIP SDDDIYDCKV EHWGLEEPVL KHWEPEIPAP MSELTETVIC ALGLSVGLVG IVVGTIFIIQ GLRSGGTSRH
11	Uniprot O19440 MHC I antigen (H2-B1)	MAQRTLFLLL AAALTMIETR AGPHSMRYFE TAVFRPGLGE PRFISVGYVD NTQFVSFDSD AENPRSEPRA PWMEQEGPEY WERETQIAKD NEQSFGWSLR NLIHYYNQSK GGFHTFQRLS GCDMGLDGRL LRGYLQFAYD GRDYITLNED LKTWMAADLV ALITRRKWEQ AGAAELYKFY LEGECVEWLR RYLELGNETL LRTDPPKAHV THHPRPAGDV TLRCWALGFY PADITLTWQL NGEELTQDME LVETRPAGDG TFQKWAAVVV PLGKEQNYTC HVYHEGLPEP LTLRWEPPPS TGSNMVNIAV LVVLGAVIII EAMVAFVLKS SRKIAILPGP AGTKGSSAS
12	Uniprot Q31191	MAPCTLLLLL AAALAPTQTR AARAAARGPV

	MHC I H2-K gene (Haplotype d) (H2-K1)	RRSGSHRAPP PGPHSLSDAD NPRFEPRAPW MEQEGPEYWE EQTQRAKSDE QWFRVSLRTA QRYYNQSKGG SHTFQRMFGC DVGSDWRLLR GYQQFAYDGR DYIALNEDLK TWTAADTAAL ITRRKWEQAG DAEYYRAYLE GECVEWLRRY LELGNETLLR TDSPKAHVTY HPRSQVDVTL RCWALGFYPA DITLTWQLNG EDLTQDMELV ETRPAGDGTF QKWAAVVVPL GKEQNYTCHV HHKGLPEPLT LRWKLPPPTV SNTVIIAVLV VLGAAIVTGA VVAFVMKMRR NTGGKGVNYA LAPGSQTSDL SLPDGKVMVH
13	Uniprot P04230 H2 Class II histocompatibility antigen E-B beta chain	MVWLPRVPCV AAVILLLTVL SPPMALVRDS RPWFLEYCKS ECHFYNGTQR VRLLERYFYN LEENLRFDSD VGEFHAVTEL GRPDAENWNS QPEFLEQKRA EVDTVCRHNY EISDKFLVRR RVEPTVTVYP TKTQPLEHHN LLVCSVSDFY PGNIEVRWFR NGKEEKTGIV STGLVRNGDW TFQTLVMLET VPQSGEVYTC QVEHPSLTDP VTVEWKAQST SAQNKMLSGV GGFVLGLLFL GAGLFIYFRN QKGQSGLQPT GLLS
14	Uniprot P04224 MHC II E-K alpha chain (underlined portion is portion used in SEQ ID NO: 30)	MATIGALVLR FFFIAVLMSS QKSWAIKEEH TIIQAEFYLL PDKRGEFMFD FDGDEIFHVD IEKSETIWRL EEFAKFASFE AQGALANIAV DKANLDVMKE RSNNTPDANV APEVTVLSRS PVNLGEPNIL ICFIDKFSPP VVNVTWLRNG RPVTEGVSET VFLPRDDHLF RKFHYLTFLP STDDFYDCEV DHWGLEEPLR KHWEFEEKTL LPETKENVVC ALGLFVGLVG IVVGIILIMK GIKKRNVVER RQGAL
15	GenBank ID: M36939.1 MHC II E-K beta chain	MWLPRVPCVAAVILLLTVLSPPVALVRDSRPWFLE YCKSECHFYNGTQRVRLLVRYFYNLEENLRFDSD VGEFRAVTELGRPDAENWNSQPEFLEQKRAEVD TVCRHNYEIFDNFLVPRRVEPTVTVYPTKTQPLEH

	(underlined portion is used in SEQ ID NO: 31, 32)	HNLLVCSVSDFYPGNIEVRWFRNGKEEKTGIVSTG LVRNGDWTFQTLVMLETVPQSGEVYTCQVEHPSL TDPVTVEWKAQSTSAQNKMLSGVGGFVLGLLFLG AGLFIYFRNQKGQSGLQPTGLLS

[0040] Referring to Table 1.1, the HLA-A (MHC I) sequence (SEQ ID NO: 1) includes the signal peptide (amino acids 1-24); amino acids 25-308 are believed to make up the extracellular region, amino acids 309-332 are believed to make up the transmembrane region, and amino acids 333-365 are believed to make up the cytoplasmic region. The HLA-B (MHC I) sequence (SEQ ID NO: 2) includes the signal peptide (amino acids 1-24); amino acids 25-308 are believed to make up the extracellular region, amino acids 309-332 are believed to make up the transmembrane region, and amino acids 333-362 are believed to make up the cytoplasmic region. The HLA-C (MHC I) sequence (SEQ ID NO: 3) includes the signal peptide (amino acids 1-24); amino acids 25-308 are believed to make up the extracellular region, amino acids 309-333 are believed to make up the transmembrane region, and amino acids 334-366 are believed to make up the cytoplasmic region. The HLA DPA1 (MHC II) sequence (SEQ ID NO: 4) includes the signal peptide (amino acids 1-28); amino acids 29-222 are believed to make up the extracellular region, amino acids 223-245 are believed to make up the transmembrane region, and amino acids 246-260 are believed to make up the cytoplasmic region. The HLA DPB1 (MHC II) sequence (SEQ ID NO: 5) includes the signal peptide (amino acids 1-29); amino acids 30-225 are believed to make up the extracellular region, amino acids 226-246 are believed to make up the transmembrane region, and amino acids 247-258 are believed to make up the cytoplasmic region. The HLA DQA1 (MHC II) sequence (SEQ ID NO: 6) includes the signal peptide (amino acids 1-23); amino acids 24-216 are believed to make up the extracellular region, amino acids 217-239 are believed to make up the transmembrane region, and amino acids 240-254 are believed to make up the cytoplasmic region. The HLA DQB1 (MHC II) sequence (SEQ ID NO: 7) includes the signal peptide (amino acids 1-32); amino acids 33-230 are believed to make up the extracellular region, amino acids 231-251 are believed to make up the transmembrane region, and amino acids 252-261 are believed to make up the cytoplasmic region. The HLA DRA (MHC II) sequence (SEQ ID NO: 8) includes the signal peptide (amino acids 1-25); amino acids 26-216 are believed to make up the extracellular region, amino acids

217-239 are believed to make up the transmembrane region, and amino acids 240-254 are believed to make up the cytoplasmic region. The HLA DRB1 (MHC II) sequence (SEQ ID NO: 9) includes the signal peptide (amino acids 1-29); amino acids 30-227 are believed to make up the extracellular region, amino acids 228-250 are believed to make up the transmembrane region, and amino acids 251-266 are believed to make up the cytoplasmic region. The MHC E-K alpha chain (SEQ ID NO: 14) includes the signal peptide (aa 1-25), the extracellular domain (aa 26-216), the transmembrane domain (aa 217-24), and a cytoplasmic portion (aa 243-255).

[0041] As previously discussed, the MHCR of the present invention comprises at least a MHC portion and a TCR portion. In some embodiments, a MHC portion comprises one or more MHC proteins (e.g., HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB1, MHC E-K alpha, MHC E-K beta, etc.), fragments thereof, or combinations thereof. For example, in some embodiments, the MHC portion comprises a fragment of any of SEQ ID NO: 1-15.

[0042] In some embodiments, the MHC portion comprises a peptide that is at least 80% identical to a MHC protein or a fragment thereof. In some embodiments, the MHC portion comprises a peptide that is at least 85% identical to a MHC protein or a fragment thereof. In some embodiments, the MHC portion comprises a peptide that is at least 90% identical to a MHC protein or a fragment thereof. In some embodiments, the MHC portion comprises a peptide that is at least 95% identical to a MHC protein or a fragment thereof. In some embodiments, the MHC portion comprises a peptide that is at least 99% identical to a MHC protein or a fragment thereof.

[0043] In some embodiments, a fragment of a MHC protein is from 10 to 25 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 50 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 100 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 150 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 200 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 250 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 10 to 350 aa in length. In some embodiments, a fragment of a MHC protein is from 25 to 50 aa in length. In some embodiments, a fragment of a MHC protein is from 25 to 100 aa in length. In some

embodiments, a fragment of a MHC protein is from 25 to 150 aa in length. In some embodiments, a fragment of a MHC protein is from 25 to 200 aa in length. In some embodiments, a fragment of a MHC protein is from 25 to 250 aa in length. In some embodiments, a fragment of a MHC protein is from 25 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 25 to 350 aa in length. In some embodiments, a fragment of a MHC protein is from 50 to 100 aa in length. In some embodiments, a fragment of a MHC protein is from 50 to 150 aa in length. In some embodiments, a fragment of a MHC protein is from 50 to 200 aa in length. In some embodiments, a fragment of a MHC protein is from 50 to 250 aa in length. In some embodiments, a fragment of a MHC protein is from 50 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 50 to 350 aa in length. In some embodiments, a fragment of a MHC protein is from 100 to 150 aa in length. In some embodiments, a fragment of a MHC protein is from 100 to 200 aa in length. In some embodiments, a fragment of a MHC protein is from 100 to 250 aa in length. In some embodiments, a fragment of a MHC protein is from 100 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 100 to 350 aa in length. In some embodiments, a fragment of a MHC protein is from 150 to 200 aa in length. In some embodiments, a fragment of a MHC protein is from 150 to 250 aa in length. In some embodiments, a fragment of a MHC protein is from 150 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 150 to 350 aa in length. In some embodiments, a fragment of a MHC protein is from 200 to 250 aa in length. In some embodiments, a fragment of a MHC protein is from 200 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 200 to 350 aa in length. In some embodiments, a fragment of a MHC protein is from 250 to 300 aa in length. In some embodiments, a fragment of a MHC protein is from 250 to 350 aa in length. In some embodiments, a fragment of a MHC protein is more than 350 aa in length.

[0044] A TCR portion may comprise one or more TCR proteins (e.g., TCRA, TCRB), one or more fragments thereof, or combinations thereof. For reference, non-limiting TCR sequences (human and mouse) are listed below in Table 2.1 and Table 2.2. The present invention is not limited to the TCR sequences in Table 2.1 and Table 2.2.

[0045] Table 2.1. Examples of Human TCR Protein Sequences

SEQ ID NO.	Description	Amino Acid Sequence
16	Uniprot P01848 T cell receptor alpha chain constant region (TRAC, TCRA)	PNIQNPDPAV YQLRDSKSSD KSVCLFTDFD SQTNVSQSKD SDVYITDKTV LDMRSMDFKS NSAVAWSNKS DFACANAFNN SIIPEDTFFP SPESSCDVKL VEKSFETDTN LNFQNLSVIG FRILLLKVAG FNLLMTLRLW SS
17	Uniprot P01850 T cell receptor beta-1 chain constant region (TRBC1)	EDLNKVFPPE VAVFEPSEAE ISHTQKATLV CLATGFFPDH VELSWWVNGK EVHSGVSTDP QPLKEQPALN DSRYCLSSRL RVSATFWQNP RNHFRCQVQF YGLSENDEWT QDRAKPVTQI VSAEAWGRAD CGFTSVSYQQ GVLSATILYE ILLGKATLYA VLVSALVLMA MVKRKDF
18	Uniprot A0A5B9 T cell receptor beta-2 chain constant region (TRBC2, TCRBC2)	DLKNVFPPEV AVFEPSEAEI SHTQKATLVC LATGFYPDHV ELSWWVNGKE VHSGVSTDPQ PLKEQPALND SRYCLSSRLR VSATFWQNPR NHFRCQVQFY GLSENDEWTQ DRAKPVTQIV SAEAWGRADC GFTSESYQQG VLSATILYEI LLGKATLYAV LVSALVLMAM VKRKDSRG
19	Uniprot B7Z8K6 T cell receptor delta chain constant region (TRDC)	SQPHTKPSVF VMKNGTNVAC LVKEFYPKDI RINLVSSKKI TEFDPAIVIS PSGKYNAVKL GKYEDSNSVT CSVQHDNKTV HSTDFEVKTD STDHVKPKET ENTKQPSKSC HKPKAIVHTE KVNMMSLTVL GLRMLFAKTV AVNFLLTAKL FFL
20	Uniprot P0CF51 T cell receptor gamma-1 chain constant region (TRGC1)	DKQLDADVSP KPTIFLPSIA ETKLQKAGTY LCLLEKFFPD VIKIHWQEKK SNTILGSQEG NTMKTNDTYM KFSWLTVPEK SLDKEHRCIV RHENNKNGVD QEIIFPPIKT DVITMDPKDN CSKDANDTLL LQLTNTSAYY MYLLLLKSV VYFAIITCCL LRRTAFCCNG EKS
21	Uniprot P03986 T cell receptor	DKQLDADVSP KPTIFLPSIA ETKLQKAGTY LCLLEKFFPD IIKIHWQEKK SNTILGSQEG

	gamma-2 chain constant region (TRGC2, TCRGC2)	NTMKTNDTYM KFSWLTVPEE SLDKEHRCIV RHENNKNGID QEIIFPPIKT DVTTVDPKDS YSKDANDVIT MDPKDNWSKD ANDTLLLQLT NTSAYYMYLL LLLKSVVYFA IITCCLLGRT AFCCNGEKS

[0046] Table 2.2. Examples of Mouse TCR Protein Sequences

SEQ ID NO.	Description	Amino Acid Sequence
22	Uniprot P01849 T cell receptor alpha chain constant region (TCRA-mouse) (underlined portion refers to sequence also used in SEQ ID NO: 30)	PYIQNPEPAV YQLKDPRSQD STLCLFTDFD SQINVPKTME SGTFITDKTV LDMKAMDSKS NGAIAWSNQT SFTCQDIFKE TNATYPSSDV PCDATLTEKS FETDMNLNFQ NLSVMGLRIL LLKVAGFNLL MTLRLWSS
23	Uniprot P01852 T cell receptor beta-1 chain constant region (TCB1-mouse)	EDLRNVTPPK VSLFEPSKAE IANKQKATLV CLARGFFPDH VELSWWVNGK EVHSGVSTDP QAYKESNYSY CLSSRLRVSA TFWHNPRNHF RCQVQFHGLS EEDKWPEGSP KPVTQNISAE AWGRADCGIT SASYQQGVLS ATILYEILLG KATLYAVLVS TLVVMAMVKR KNS
24	Uniprot P01851 T cell receptor beta-2 chain constant region (TCB2-mouse) (underlined portion refers to sequence used in SEQ ID NO: 31, 32)	EDLRNVTPPK VSLFEPSKAE IANKQKATLV CLARGFFPDH VELSWWVNGK EVHSGVSTDP QAYKESNYSY CLSSRLRVSA TFWHNPRNHF RCQVQFHGLS EEDKWPEGSP KPVTQNISAE AWGRADCGIT SASYHQGVLS ATILYEILLG KATLYAVLVS GLVLMAMVKK KNS
25	Uniprot P01853 T cell receptor gamma chain constant region C10.5 (TCC1-mouse)	DKRLDADISP KPTIFLPSVA ETNLHKTGTY LCLLEKFFPD VIRVYWKEKN GNTILDSQEG DTLKTKGTYM KFSWLTVPER AMGKEHSCIV KHENNKGGAD QEIFFPSIKK VATTCWQDKN

		DVLQFQFTST SAYYTYLLLL LKSVIYLAII SFSLLRRTSV CGNEKKS
26	Uniprot P03985 T cell receptor gamma chain constant region C7.5 (TCC2-mouse)	DKKLDADISP KPTIFLPSVA ETNLHKTGTY LCVLEKFFPD VIRVYWKEKK GNTILDSQEG DMLKTNDTYM KFSWLTVPER SMGKEHRCIV KHENNKGGAD QEIFFPTIKK VAVSTKPTTC WQDKNDVLQL QFTITSAYYT YLLLLLKSVI YLAIISFSLL RRTSVCCNEK KS
27	Uniprot P06334 T cell receptor gamma chain constant region DFL12 (TCC3-mouse)	PSDKRLDADI SPKPTIFLPS VAETNLHKTG TYLCILEKFF PDVIRVYWKD KNGNTILDSQ EGDTLKTKGT YMKFSWLTVP ERSMGKEHRC IVKHENNKGG ADQEIFFPSI KKVATTCWQD KNDVLQLQFM STSAYYTYLL LLLKSVIYLA IISFSLLRRT SVCCNEKRS
28	Uniprot P06335 T cell receptor gamma chain constant region 5/10-13 (TCC4-mouse)	DKRTDSDFSP KPTIFLPSAA ETNLHKAGTY LCLLEKFFPK VIRVYWKEKD GEKILESQEG NTIKTNDRYM KFSWLTVTED SMAKEHSCIV KHENNKRGVD QEILFPPIGK AFTTINVNPR DSVLRHENVN NATDLEDCMK GRKDMLQLQV TTTYAFYTYL ILFFKSMVHL AFVVFCLFRR AAMSCDDQRS

[0047] Referring to the TRAC protein (SEQ ID NO: 16) in Table 2, amino acids 118-137 are believed to make up the transmembrane domain, and amino acids 138-142 are believed to make up the cytoplasmic domain. Referring to the TRBC1 protein (SEQ ID NO: 17) in Table 2, amino acids 151-171 are believed to make up the transmembrane domain. Referring to the TRBC2 protein (SEQ ID NO: 18) in Table 2, amino acids 145-167 are believed to make up the transmembrane domain. Referring to the TRDC protein (SEQ ID NO: 19) in Table 2, amino acids 130-152 are believed to make up the transmembrane domain. Referring to the TRGC1 protein (SEQ ID NO: 20) in Table 2, amino acids 139-161 are believed to make up the transmembrane domain. Referring to the TRGC2 protein (SEQ ID NO: 21) in Table 2, amino acids 157-177 are believed to make up the transmembrane domain, and amino acids 178-189 are believed to make up

the cytoplasmic domain.

[0048] As previously discussed, the MHCR of the present invention comprises at least a MHC portion and a TCR portion. In some embodiments, a TCR portion comprises one or more TCR proteins (e.g., TRAC, TRBC1, TRBC2, TRDC, TRCG1, TRCG2, TCRA-mouse, TCB1-mouse, TCB2-mouse, TCC1-mouse, TCC2-mouse, TCC3 mouse, TCC4 mouse, etc.), fragments thereof, or combinations thereof. For example, in some embodiments, the TCR portion comprises a fragment of any of SEQ ID NO: 16-28. (In some embodiments, the fragment is from 5 to 10 aa in length. In some embodiments, the fragment is from 10 to 20 aa in length, in some embodiments, the fragment is from 10 to 30 aa in length. IN some embodiments, the fragment is from 10 to 40 aa in length. In some embodiments, the fragment is from 10 to 50 aa in length, etc.

[0049] In some embodiments, the TCR portion comprises a peptide that is at least 80% identical to a TCR protein (e.g., any of SEQ ID NO: 16-28), or a fragment thereof. In some embodiments, the TCR portion comprises a peptide that is at least 85% identical to a TCR protein (e.g., any of SEQ ID NO: 16-28), or a fragment thereof. In some embodiments, the TCR portion comprises a peptide that is at least 90% identical to a TCR protein (e.g., any of SEQ ID NO: 16-28), or a fragment thereof. In some embodiments, the TCR portion comprises a peptide that is at least 95% identical to a TCR protein (e.g., any of SEQ ID NO: 16-28), or a fragment thereof. In some embodiments, the TCR portion comprises a peptide that is at least 99% identical to a TCR protein (e.g., any of SEQ ID NO: 16-28), or a fragment thereof.

[0050] In some embodiments, a fragment of a TCR protein is from 10 to 25 aa in length. In some embodiments, a fragment of a TCR protein is from 10 to 50 aa in length. In some embodiments, a fragment of a TCR protein is from 10 to 100 aa in length. In some embodiments, a fragment of a TCR protein is from 10 to 150 aa in length. In some embodiments, a fragment of a TCR protein is from 25 to 50 aa in length. In some embodiments, a fragment of a TCR protein is from 25 to 100 aa in length. In some embodiments, a fragment of a TCR protein is from 25 to 150 aa in length. In some embodiments, a fragment of a TCR protein is from 50 to 100 aa in length. In some embodiments, a fragment of a TCR protein is from 50 to 150 aa in length. In some embodiments, a fragment of a TCR protein is from 100 to 150 aa in length. In some embodiments, a fragment of a TCR protein is more than 150 aa in length.

[0051] In some embodiments, the MHCR comprises a peptide antigen. Any appropriate peptide antigen may be used. The peptide antigen in the pMHCR complex directs the specificity of the pMHCR molecule, therefore the pMHCR molecule will be specific for T cells with TCRs that are specific for that peptide antigen/pMHCR. A non-limiting example of a peptide antigen that may be used with the MHCR is moth cytochrome c peptide (aa 88-103, ANERADLIAYLKQATK (SEQ ID NO: 29)). The peptide antigens used in the Examples (see below) are peptides commonly used as model antigens in mouse models. Any appropriate peptide antigen may be used, and the present invention is not limited to the peptide antigens disclosed herein. For example, in some embodiments, the peptide antigen comprises any immunodominant peptide antigen identified to bind a class I or class II MHC. In some embodiments, the peptide antigen comprises any immunodominant peptide antigen identified to bind a class I or class II MHC and elicit a response. A response may include but is not limited to an autoimmune response, an allergic response, an asthma response, or an inappropriate Treg response. The peptide antigen may be any appropriate length.

[0052] In some embodiments, the MHCR comprises at least a portion of a MHC molecule that allows for binding to an appropriate TCR. In some embodiments, the MHCR comprises at least a portion of a MHC molecule that allows for binding to an appropriate TCR and at least a portion of a TCR molecule (e.g., a portion of a TCR molecule that allows for appropriate signaling and/or complexing subunits such as CD3 subunits). In some embodiments, the MHCR comprises a transmembrane domain that is at least partially derived from (i) a MHC molecule, (ii) a TCR molecule, or (iii) both the MHC molecule and TCR molecule. In some embodiments, the MHCR comprises a transmembrane domain, wherein a portion (or all) of the transmembrane domain is not derived from a MHC molecule or a TCR molecule. In some embodiments, the MHCR comprises an extracellular domain that is at least partially derived from (i) a MHC molecule, (ii) a TCR molecule, or (iii) both the MHC molecule and TCR molecule. In some embodiments, the MHCR comprises an extracellular domain, wherein a portion of the extracellular domain is not derived from a MHC molecule or a TCR molecule.

[0053] As an example, in some embodiments, the MHCR comprises at least a portion of the extracellular domain of a MHC molecule (e.g., the extracellular domain of HLA-DRA) and at least a portion of the transmembrane domain of a TCR molecule and at least a

portion of the cytoplasmic domain of a TCR molecule. As another example, in some embodiments, the MHCR comprises at least a portion of the extracellular domain of a TCR molecule.

[0054] The present invention also features redirected cells, such as redirected T cells, expressing MHCRs of the present invention, e.g., as described above. Without wishing to limit the present invention to any theory or mechanism, the MHCRs are generally adapted to recognize and bind to appropriate (specific) TCRs. In some embodiments, the MHCR is expressed in a CD8+ T cell (e.g., a cytotoxic T cell, T_C cells, CTLs). In some embodiments, the MHCR is expressed in a CD4+ T cell (e.g., a T helper cell, T_H cell or a regulatory T cell (Treg cell)). The present invention is not limited to the expression of MHCRs in T cells, nor is the present invention limited to expression of MHCRs in CD8+ or CD4+ T cells, e.g., the MHCRs may be expressed in CD8+/CD4+ thymocytes, γδT cells, NK cells, NK T cells, etc. In some embodiments, the MHCR of the redirected T cell complexes or is adapted to complex with CD3 subunits (e.g., forming a MHCR-CD3 complex).

[0055] In some embodiments, the MHCR comprises a MHC portion derived from an extracellular portion of a MHC protein and a TCR portion derived from a transmembrane domain of a TCR protein. In some embodiments, the MHC portion and TCR portion are directly linked. In some embodiments, the MHC portion and TCR portion are separated by a linker. In some embodiments, the linker comprises a glycine-rich linker.

[0056] The present invention is not limited to the MHC portions and TCR portions described herein. For example, the MHC portion may comprise any MHC peptide, e.g., an extracellular domain (or a portion thereof) of any MHC peptide. The TCR portion may comprise any TCR peptide, e.g., a transmembrane domain (or portion thereof) of any TCR peptide. Further, the present invention is not limited to antigens, signaling molecules, and cell surface receptor ligands described herein, e.g., the present invention may be applicable to a wide range of MHC molecules, TCR molecules, antigens, signaling moleucles cell surface receptor ligands, etc.

Surrogate Coreceptors (SCRs)

[0057] The present invention also features chimeric surrogate coreceptors (SCR), e.g., receptors that recruit signaling molecules (e.g., kinases such as but not limited to Src

kinases (e.g., Lck), phosphatases, etc.). In some embodiments, the SRCs recruit signaling molecules (e.g., kinases) to the MHCR and/or CD3 subunits. The present invention also features cells expressing a SCR. In some embodiments, redirected cells, e.g., redirected T cells, express both a MHCR and a SCR. In some embodiments, cells express more than one type of SCR. Without wishing to limit the present invention to any theory or mechanism, it is believed that certain SCRs may enhance signaling through the pMHCR-CD3 complex.

[0058] In some embodiments, the SCR comprises a cell surface receptor ligand (e.g., T cell surface receptor ligand) fused to a signaling molecule (e.g., kinase (e.g., Lck or other appropriate kinase), phosphatase, etc.). In some embodiments, the cell surface receptor ligand and the kinase are separated by a linker, e.g., a peptide linker or any other appropriate linker. The signaling molecule is not limited to a kinase or a phosphatase.

[0059] In some embodiments, the cell surface receptor ligand (e.g., T cell surface receptor ligand) comprises CD80, CD86, fragments thereof, or combinations thereof. The present invention is not limited to CD80 and CD86; any other appropriate cell surface receptor ligand (or a fragment thereof) may be used. For example, in some embodiments, the cell surface receptor ligand comprises a CD28 ligand, a CTLA-4 ligand, an ICOS ligand, an OX4O ligand, a PD-1 ligand (e.g., PD-1L), a CD2 ligand, etc.

[0060] As an example, in some embodiments, when a T cell is expressing a pMHCR (a MHCR with a peptide antigen), the pMHCR may complex with CD3 subunits, forming a pMHCR-CD3 complex. If the cell is also expressing a CD80-Lck SCR, then when the pMHCR binds a TCR on a target T cell, the CD80-Lck may also bind to CD28 on the same target T cell. Without wishing to limit the present invention to any theory or mechanism, it is believed that then the CD80-Lck SCR should recruit Lck to the pMHCR-CD3 complex to phosphorylate the pMHCR-CD3 ITAMs for robust signaling.

[0061] In some embodiments, the SCR is engineered (e.g., a particular cell surface receptor ligand of the SCR is selected) to target a specific set of target cells. For example, T follicular helper cells express a molecule called PD-1 and these cells provide help to B cells to make autoantibodies in autoimmune diseases such as Lupus. The ligand for PD-1 is PD-1L, so a SCR comprising PD-1L and Lck may be co-expressed

with a pMHCR recognized by the TCR of the T follicular helper cell. This may allow for targeting of this specific T follicular helper cell population.

[0062] The present invention also features methods of use of said MHCRs, SCRs, and/or said redirected cells, for example for immunotherapy. In some embodiments, the redirected cells may eliminate autoreactive T cells, regulatory T cells (Tregs) that protect tumor cells by suppressing anti-tumor T cell responses, or any other appropriate T cell. For example, in some embodiments, the MHCR is an auto-antigen MHCR, and the MHCR's target is an autoreactive T cell.

EXAMPLES

[0063] **Example 1: Redirected T cells targeting CD4 T Helper Cells.** Example 1 describes a non-limiting experimental approach to target CD4 T cells. A prototype pMHCR was engineered with a peptide antigen: the moth cytochrome c peptide (SEQ ID NO: 29) was fused to the mouse class II MHC I-E^k (MCC:I-E^k; e.g., see SEQ ID NO: 31). This pMHCR was expressed (e.g., retrovirally expressed) in T cell hybridomas. It was determined that this pMHCR (e.g., pMHCR-CD3 complex) was expressed on the surface of T cell hybridomas (see FIG. 4). IL-2 production was induced after interactions with cognate TCRs (e.g., 5c.c7, 2B4), yet an irrelevant peptide (control peptide antigen) in the pMHCR-CD3 complex rendered it non-stimulatory (data not shown).

[0064] Lck fusions were generated with known ligands for T cell surface receptors. For example, all T cells express CD28. Lck fusions with CD28 ligands (e.g., CD80, CD86) were engineered to generate surrogate coreceptors (SCRs), e.g., CD80-Lck (see SEQ ID NO: 33, SEQ ID NO: 38), e.g., CD86-Lck (see SEQ ID NO: 34, SEQ ID NO: 39). When the pMHCR-CD3 complex was co-expressed with SCR CD80-Lck in hybridomas, these cells produced significantly more IL-2 in response to cells expressing the 2B4 TCR ligand + CD28 than they did in response to cells expressing only the 2B4 TCR ligand (see FIG. 5). This suggested that signaling through the pMHCR-CD3 complex could be augmented through the use of a SCR.

[0065] MCC:IE^k pMHCR-CD3 and the SCR CD80-Lck or HB:IE^k pMHCR-CD3 (e.g., see SEQ ID NO: 32) and the SCR CD80-Lck were expressed in *in vitro* differentiated CD8 cytotoxic T cells (CTLs) and their ability to kill 5c.c7 TCR transgenic CD4 T cells expressing the TCR specific for the MCC:IE^k pMHCR was evaluated. Surface

expression of the pMHCRs on the redirected CTLs was observed, suggesting that these chimeric receptor modules compete with the endogenous TCR for assembly with the endogenous CD3 subunits (data not shown). CTLs expressing the MCC:IE^k pMHCR robustly killed the target CD4 T cells while those expressing the null HB:IE^k pMHCR did not (see FIG. 6). This suggests that CD8 T cells can be redirected to target and eliminate antigen-specific CD4 T cells.

[0066] **Example 2: Redirected T cells targeting CD4 T Helper Cells in Allergic Asthma.** Example 2 describes a non-limiting experimental approach to target CD4 T helper cells involved in allergic asthma, e.g., to help eliminate naïve Der p 1-specific CD4 T cells from the repertoire prior to House Dust Mite (HDM) sensitization. Without wishing to limit the present invention to any theory or mechanism, it is believed that eliminating allergen-specific CD4 T cells from the repertoire may help prevent the onset of T_H2 immunity upon HDM sensitization.

[0067] A pMHCR (pMHCR-CD3 complex) will be retrovirally expressed in *in vitro* activated CTLs. The pMHCR will bear a pMHCR comprising either the immunodominant HDM-derived Der p 1 epitope (aa117-127) in the context of I-A^b (Derp1:IA^b) or the immunodominant West Nile Virus peptide from the envelope protein (aa641-655) in the context of I-A^b (E641:IA^b). The E641:IA^b pMHCR cells will serve as a non-specific control population.

[0068] The *in vitro* activated CTLs will also be transduced with a CD80-Lck SCR to enhance signaling. These redirected CTLs will then be transferred intravenously into C57Bl/6 mice to target and eliminate Derp1:IA^b- or E641:IA^b-specific naïve CD4 T cells from the endogenous repertoire. After a certain length of time, e.g., 1 week, the elimination of antigen-specific CD4 T cells will be evaluated. This will be performed via tetramer enrichment experiments using a Derp1:IA^b tetramer and a E641:IA^b tetramer. The presence of the redirected CD8 T cells will also be assessed by flow cytometry by gating on CD3$^+$CD8$^+$IA^{b+} T cells since mouse T cells do not express class II MHC.

[0069] After determining if the redirected CTLs eliminate the target population, mice that received redirected CTLs one-week prior will be sensitized with HDM (e.g., intranasally, e.g., with HDM extracts). This will be done even if endogenous CD4 T cells specific for Derp1:IA^b are detected, but only if redirected T cells are still present in the mice. This

may help to determine if activation of the CD4 T cells made them more susceptible to targeting by the redirected CTLs.

[0070] **Example 3: Redirected T cells targeting CD4 T Helper Cells in Lungs After Sensitization.** Example 3 describes a non-limiting experimental approach to target CD4 T helper cells in lungs of HDM-sensitized mice. Without wishing to limit the present invention to any theory or mechanism, it is believed that eliminating allergen-specific CD4 T cells from the lungs of HDM-sensitized mice may help attenuate T_H2 immunity.

[0071] Der p 1-specific CD4 T cells will be targeted similarly to Example 2, but only after HDM sensitization. In brief, mice will be sensitized with HDM according to the protocol described above. They will then receive redirected Derp1:IA^b or E641:IA^b pMHCR-CD3 CTLs on day 14. Various surrogate co-receptors will be employed to explore the efficacy of the technology and approach. For example, the CD80-Lck fusion SCR will be used, as well as others, e.g., a TIM-4-Lck SCR (since the TIM-1 expressed on CD4 T cells is genetically linked with asthma and this combination for targeting might enhance effectiveness). One week after transfer of redirected CTLs, cytokine and cellular analysis will be performed as described above in Example 2 so as to assess the impact of these cells on the lung cytokine milieu and cellularity. The status of the redirected CTLs will also be evaluated.

[0072] **Example 4: Attenuation of Der p 1-specific CD4 T cell function *in situ*.** Example 4 describes a non-limiting experimental approach to redirect Tregs against Der p 1-specific CD4 T cells. Without wishing to limit the present invention to any theory or mechanism, it is believed that this may help attenuate function of said CD4 T cells and help diminish T_H2 immunity.

[0073] *In vitro* generated induced Tregs (iTregs) expressing a MHCR will be tested for efficacy in reducing HDM-induced airway hypersensitivity. Induced Tregs (iTregs) will be generated *in vitro* and transduced with pMHCR and SCRs as described in Examples 2 and 3 above. These cells will then either be transferred prior to HDM sensitization as in Example 2 or after sensitization as in Example 3. Evaluation of the lung cytokine milieu and cellularity will then be performed as described above.

[0074] Table 3 shows examples of protein sequences for reagents the above examples. Table 4 shows the nucleotide sequences for the proteins in Table 3. Note that in SEQ ID

NO: 30, a portion is derived from SEQ ID NO: 14 and a portion is derived from SEQ ID NO: 22. In SEQ ID NO: 31, a portion is derived from SEQ ID NO: 15, a portion is derived from SEQ ID NO: 23, and a portion is derived from SEQ ID NO: 29 (and other residues may correspond to a glycine-rich linking region). In SEQ ID NO: 32, a portion is derived from SEQ ID NO: 15 and a portion is derived from SEQ ID NO: 23 (and other residues may correspond to a glycine-rich linking region).

[0075] Table 3. Peptide sequences for reagents in Examples.

SEQ ID NO.	Description	Amino Acid Sequence
30	I-E^kα-TCRα Note: underlined portion is from SEQ ID NO: 14 (MHC portion), bold portion is from SEQ ID NO: 22 (TCR portion)	MATIGALLLRFFFIAVLMSSQKSWAIKEEHTIIQAEFY LLPDKRGEFMFDFDGDEIFHVDIEKSETIWRLEEFA KFASFEAQGALANIAVDKANLDVMKERSNNTPDAN VAPEVTVLSRSPVNLGEPNILICFIDKFSPPVVNVTW FRNGRPVTEGVSETVFLPRDDHLFRKFHYLTFLPS TDDFYDCEVDHWGLEEPLRKHWEFEEKTLLPETK E**CDATLTEKSFETDMNLNFQNLSVMGLRILLLKVA GFNLLMTLRLWSS**
31	MCC:I-E^kβ-TCRβ (note: italic portion shows peptide antigen sequence, underlined portion is from SEQ ID NO: 15 (MHC portion), and bold portion is from SEQ ID NO: 24 (TCR portion)	MVWLPRVPCVAAVILLLTVLSPPVALVRDSGS*ANE RADLIAYLKQAT*KEFRSGGGGSLVPRGSGGGGSV DRPWFLEYCKSECHFYNGTQRVRLLVRYFYNLEE NLRFDSDVGEFRAVTELGRPDAENWNSQPEFLEQ KRAEVDTVCRHNYEIFDNFLVPRRVEPTVTVYPTKT QPLEHHNLLVCSVSDFYPGNIEVRWFRNGKEEKTG IVSTGLVRNGDWTFQTLVMLETVPQSGEVYTCQVE HPSLTDPVTVEWKAQSTSAQNK**CGITSASYHQGV LSATILYEILLGKATLYAVLVSGLVLMAMVKKKNS** AAA
32	HB:I-E^kβ-TCRβ Note: italic portion shows peptide antigen sequence, underlined portion is	MVWLPRVPCVAAVILLLTVLSPPVALVRDSGSGKK VITAFNEGLKEFRSGGGGSLVPRGSGGGGSVDRP WFLEYCKSECHFYNGTQRVRLLVRYFYNLEENLRF DSDVGEFRAVTELGRPDAENWNSQPEFLEQKRAE VDTVCRHNYEIFDNFLVPRRVEPTVTVYPTKTQPLE

	from SEQ ID NO: 15 (MHC portion), and bold portion is from SEQ ID NO: 24 (TCR portion)	HHNLLVCSVSDFYPGNIEVRWFRNGKEEKTGIVST GLVRNGDWTFQTLVMLETVPQSGEVYTCQVEHPS LTDPVTVEWKAQSTSAQNK**CGITSASYHQGVLSAT ILYEILLGKATLYAVLVSGLVLMAMVKKKNS**AAA
33	CD80-Lck (mCD80-mLck fusion)	MACNCQLMQDTPLLKFPCPRLILLFVLLIRLSQVSS DVDEQLSKSVKDKVLLPCRYNSPHEDESEDRIYWQ KHDKVVLSVIAGKLKVWPEYKNRTLYDNTTYSLIILG LVLSDRGTYSCVVQKKERGTYEVKHLALVKLSIKAD FSTPNITESGNPSADTKRITCFASGGFPKPRFSWLE NGRELPGINTTISQDPESELYTISSQLDFNTTRNHTI KCLIKYGDAHVSEDFTWEKPPEDPPDSKNTLVLFG AGFGAVITVVVIVVIIKCFCKHRSCFRRNEASRETNN SLTFGPEEALAEQTVFLTTSHYPIVPLDSKISLPIRN GSEVRDPLVTYEGSLPPASPLQDNLVIALHSYEPSH DGDLGFEKGEQLRILEQSGEWWKAQSLTTGQEGF IPFNFVAKANSLEPEPWFFKNLSRKDAERQLLAPG NTHGSFLIRESESTAGSFSLSVRDFDQNQGEVVKH YKIRNLDNGGFYISPRITFPGLHDLVRHYTNASDGL CTKLSRPCQTQKPQKPWWEDEWEVPRETLKLVER LGAGQFGEVWMGYYNGHTKVAVKSLKQGSMSPD AFLAEANLMKQLQHPRLVRLYAVVTQEPIYIITEYME NGSLVDFLKTPSGIKLNVNKLLDMAAQIAEGMAFIE EQNYIHRDLRAANILVSDTLSCKIADFGLARLIEDNE YTAREGAKFPIKWTAPEAINYGTFTIKSDVWSFGILL TEIVTHGRIPYPGMTNPEVIQNLERGYRMVRPDNC PEELYHLMMLCWKERPEDRPTFDYLRSVLDDFFTA TEGQYQPQPGT
34	CD86-Lck (mCD86-mLck fusion)	MDPRCTMGLAILIFVTVLLISDAVSVETQAYFNGTAY LPCPFTKAQNISLSELVVFWQDQQKLVLYEHYLGT EKLDSVNAKYLGRTSFDRNNWTLRLHNVQIKDMG SYDCFIQKKPPTGSIILQQTLTELSVIANFSEPEIKLA QNVTGNSGINLTCTSKQGHPKPKKMYFLITNSTNE

		YGDNMQISQDNVTELFSISNSLSLSFPDGVWHMTV VCVLETESMKISSKPLNFTQEFPSPQTYWKEITASV TVALLLVMLLIIVCHKKPNQPSRPSNTASKLERDSN ADRETINLKELEPQIASAKPNAECTSHYPIVPLDSKI SLPIRNGSEVRDPLVTYEGSLPPASPLQDNLVIALH SYEPSHDGDLGFEKGEQLRILEQSGEWWKAQSLT TGQEGFIPFNFVAKANSLEPEPWFFKNLSRKDAER QLLAPGNTHGSFLIRESESTAGSFSLSVRDFDQNQ GEVVKHYKIRNLDNGGFYISPRITFPGLHDLVRHYT NASDGLCTKLSRPCQTQKPQKPWWEDEWEVPRE TLKLVERLGAGQFGEVWMGYYNGHTKVAVKSLKQ GSMSPDAFLAEANLMKQLQHPRLVRLYAVVTQEPI YIITEYMENGSLVDFLKTPSGIKLNVNKLLDMAAQIA EGMAFIEEQNYIHRDLRAANILVSDTLSCKIADFGLA RLIEDNEYTAREGAKFPIKWTAPEAINYGTFTIKSDV WSFGILLTEIVTHGRIPYPGMTNPEVIQNLERGYRM VRPDNCPEELYHLMMLCWKERPEDRPTFDYLRSV LDDFFTATEGQYQPQPGT

[0076] Table 4. Examples of DNA sequences for encoding the proteins in Table 3.

SEQ ID NO.	Description	Gene Sequence
35	I-E^kα-TCRα fusion	aataagcttctcgagcgccaccATGGCCACAATTGGAGCCCTGCTGTTAAGATTT TTCTTCATTGCTGTTCTGATGAGCTCCCAGAAGTCATGGGCTATCAAA GAGGAACACACCATCATCCAGGCGGAGTTCTATCTTTTACCAGACAAA CGTGGAGAGTTTATGTTTGACTTTGACGGCGATGAGATTTTCCATGTA GACATTGAAAAGTCAGAGACCATCTGGAGACTTGAAGAATTTGCAAAG TTTGCCAGCTTTGAGGCTCAGGGTGCACTGGCTAATATAGCTGTGGAC AAAGCTAACCTGGATGTCATGAAAGAGCGTTCCAACAACACTCCAGAT GCCAACGTGGCCCCAGAGGTGACTGTACTCTCCAGAAGCCCTGTGAA CCTGGGAGAGCCCAACATCCTCATCTGTTTCATTGACAAGTTCTCCCC TCCAGTGGTCAATGTCACCTGGTTCCGGAATGGACGGCCTGTCACCG AAGGCGTGTCAGAGACAGTGTTTCTCCCGAGGGACGATCACCTCTTC CGCAAATTCCACTATCTGACCTTCCTGCCCTCCACAGATGATTTCTATG ACTGTGAGGTGGATCACTGGGGTTTGGAGGAGCCTCTGCGGAAGCAC

		TGGGAGTTTGAAGAGAAAACCCTCCTCCCAGAAACTAAAGAGtgtgatgcc acgttgaccgagaaaaGCTTTGAAACAGATATgaacctaaactttcaaaacctgtcaGTT ATGGGACTCCGAATCCtcctgctgaaagtagcgggatttaacCTGCTCATGACGCT gaggctgtggtccagttgaggatccgcta
36	MCC:I-E^{k}β-TCRβ fusion	aatCTCGAGCGCCACCATGGTGTGGCTCCCCAGAGTTCCCTGTGTGGC AGCTGTGATCCTGTTGCTGACAGTGCTGAGCCCTCCAGTGGCTTTGG TCAGAGACTCCGGATCCGCCAACGAGAGGGCCGACCTGATCGCCTAC CTGAAGCAGGCCACCAAGGAATTCAGATCCGGAGGCGGAGGCTCCCT GGTGCCTCGGGGCTCCGGAGGCGGAGGCTCCGTCGACAGACCATGG TTTTTGGAATACTGTAAATCTGAGTGTCATTTCTACAACGGGACGCAG CGCGTGCGGCTTCTGGTAAGATACTTCTACAACCTGGAGGAGAACCT GCGCTTCGACAGCGACGTGGGCGAGTTCCGCGCGGTGACCGAGCTG GGGCGGCCAGACGCCGAGAACTGGAACAGCCAGCCGGAGTTCCTGG AGCAAAAGCGGGCCGAGGTGGACACGGTGTGCAGACACAACTATGA GATCTTCGATAACTTCCTTGTGCCGCGGAGAGTTGAGCCTACGGTGA CTGTGTACCCCACAAAGACGCAGCCCCTGGAACACCACAACCTCCTG GTCTGCTCTGTGAGTGACTTCTACCCTGGCAACATTGAAGTCAGATGG TTCCGGAATGGCAAGGAGGAGAAAACAGGAATTGTGTCCACGGGCCT GGTCCGAAATGGAGACTGGACCTTCCAGACACTGGTGATGCTGGAGA CGGTTCCTCAGAGTGGAGAGGTTTACACCTGCCAGGTGGAGCATCCC AGCCTGACCGACCCTGTCACGGTCGAGTGGAAAGCACAGTCCACATC TGCACAGAACAAGtgtggaatcactagtgcatcctatcatcaggggttctgtctgcaaccatcct ctatgagatcctactggggaaggccaccctatatgctgtgctggtcagtggcctagtgctgatgGCCA TGGTCAAGAAAAAAAATTCCgcggccgcatgatgagatctgagctccatagaggcg
37	HB:I-E^{k}β-TCRβ fusion	aatCTCGAGCGCCACCATGGTGTGGCTCCCCAGAGTTCCCTGTGTGGC AGCTGTGATCCTGTTGCTGACAGTGCTGAGCCCTCCAGTGGCTTTGG TCAGAGACTCCGGATCCGGCAAGAAGGTGATCACCGCCTTCAACGAG GGCCTGAAGGAATTCAGATCCGGAGGCGGAGGCTCCCTGGTGCCTC GGGGCTCCGGAGGCGGAGGCTCCGTCGACAGACCATGGTTTTTGGA ATACTGTAAATCTGAGTGTCATTTCTACAACGGGACGCAGCGCGTGCG GCTTCTGGTAAGATACTTCTACAACCTGGAGGAGAACCTGCGCTTCGA CAGCGACGTGGGCGAGTTCCGCGCGGTGACCGAGCTGGGGCGGCC AGACGCCGAGAACTGGAACAGCCAGCCGGAGTTCCTGGAGCAAAAG CGGGCCGAGGTGGACACGGTGTGCAGACACAACTATGAGATCTTCGA TAACTTCCTTGTGCCGCGGAGAGTTGAGCCTACGGTGACTGTGTACC CCACAAAGACGCAGCCCCTGGAACACCACAACCTCCTGGTCTGCTCT GTGAGTGACTTCTACCCTGGCAACATTGAAGTCAGATGGTTCCGGAAT GGCAAGGAGGAGAAAACAGGAATTGTGTCCACGGGCCTGGTCCGAAA TGGAGACTGGACCTTCCAGACACTGGTGATGCTGGAGACGGTTCCTC AGAGTGGAGAGGTTTACACCTGCCAGGTGGAGCATCCCAGCCTGACC

		GACCCTGTCACGGTCGAGTGGAAAGCACAGTCCACATCTGCACAGAA CAAGtgtggaatcactagtgcatcctatcatcaggggttctgtctgcaaccatcctctatgagatccta ctggggaaggccaccctatatgctgtgctggtcagtggcctagtgctgatgGCCATGGTCAAGA AAAAAAATTCCgcggccgcatgatgagatctgagctccatagaggcg
38	CD80-Lck (mCD80- mLck fusion)	acgtctagatacctcgaggccaccATGGCTTGCAATTGTCAGttgatgcaggatacaccac tcctcaagtttccatgtccaaggctcattcttctctttgtgctgctgattcgtctttcacaagtgtcttcagatgttg atgaacaactgtccaagtcagtgaaagataaggtattgctgccttgccgttacaactctcctcatgaagat gagtctgaagaccgaatctactggcaaaaacatgacaaagtggtgctgtctgtcattgctgggaaacta aaagtgtggcccgagtataagaaccggactttatatgacaacactacctactctcttatcatcctgggcct ggtcctttcagaccggggcacatacagctgtgtcgttcaaaagaaggaaagaggaacgtatgaagtta aacacttggctttagtaaagttgtccatcaaagctgacttctctaccccccaacataactgagtctggaaac ccatctgcagacactaaaaggattacctgctttgcttccgggggtttcccaaagcctcgcttctcttggttgg aaaatggaagagaattacctggcatcaatacgacaatttcccaggatcctgaatctgaattgtacaccat tagtagccaactagatttcaatacgactcgcaaccacaccattaagtgtctcattaaatatggagatgctc acgtgtcagaggacttcacctgggaaaaaccccccagaagaccctcctgatagcaagaacacacttgt gctctttggggcaggattcggcgcagtaataacagtcgtcgtcatcgttgtcatcatcaaatgcttctgtaa gcacagaagctgtttcagaagaaatgaggcaagcagagaaacaaacaacagccttaccttcgggcc tgaagaagcattagctGAACAGACCGTCTTCCTTaccactagtCACTATCCCATAG TCccactggacagcaagatctcgctgcccatccggaatggctctgaagtgcgggacccactggtcac ctatgagggatctctcccaccagcatccccgctgcaagacaacctggttatcgccctgcacagttatgag ccctcccatgatggagacttgggctttgagaagggtgaacagctccgaatcctggagcagagcggtga gtggtggaaggctcagtccctgacgactggccaagaaggcttcattcccttcaacttcgtggcgaaagc aaacagcctggagcctgaaccttggttcttcaagaatctgagccgtaaggacgccgagcggcagcttttt ggcgcccgggaacacgcatggatccttcctgatccgggaaagcgaaagcactgcggggtcctttccc tgtcggtcagagacttcgaccagaaccagggagaagtggtgaaacattacaagatccgtaacctaga caacggtggcttctacatctcccctcgtatcacttttcccggattgcacgatctagtccgccattacaccaa cgcctctgatgggctgtgcacaaagttgagccgtccttgccagacccagaagccccagaaaccatggt gggaggacgaatgggaagttcccagggaaacactgaagttggtggagcggctgggagctggccagt tcggggaagtgtggatggggtactacaacggacacacgaaggtggcggtgaagagtctgaaacaag ggagcatgtcccccgacgccttcctggctgaggctaacctcatgaagcagctgcagcacccgcggcta gtccggctttatgcagtggtcacccaggaacccatctacatcatcacggaatacatggagaacgggag cctagtagattttctcaagactcccctcgggcatcaagttgaatgtcaacaaacttttggacatggcagccc agattgcagagggcatggcgttcatcgaagaacagaattacatccatcgggacctgcgcgccgccaa catcctggtgtctgacacgctgagctgcaagattgcagactttggcctggcgcgcctcattgaggacaat gagtacacggcccgggaggggccaaatttcccattaagtggacagcaccagaagccattaactatg ggaccttcaccatcaagtcagacgtgtggtccttcgggatcttgcttacagagatcgtcacccacggtcg aatcccttacccaggaatgaccaaccctgaagtcattcagaacctggagagaggctaccgcatggtga gacctgacaactgtccggaagagctgtaccacctcatgatgctgtgctggaaggagcgcccagagga ccggcccacgtttgactaccttcggagtgttctggatgacttcttcacagccacagagggcCAGTAC CAGCCCCAGCCTggtacctagtgagaattctacatg

39	CD86-Lck (mCD86-mLck fusion)	tactctagatacctcgaggccaccATGGACCCCAGATGCACCatgggcttggcaatccttat ctttgtgacagtcttgctgatctcagatgctgtttccgtggagacgcaagcttatttcaatgggactgcatatc tgccgtgcccatttacaaaggctcaaaacataagcctgagtgagctggtagtattttggcaggaccagc aaaagttggttctgtacgagcactatttgggcacagagaaacttgatagtgtgaatgccaagtacctggg ccgcacgagctttgacaggaacaactggactctacgacttcacaatgttcagatcaaggacatgggctc gtatgattgttttatacaaaaaaagccacccacaggatcaattatcctccaacagacattaacagaactg tcagtgatcgccaacttcagtgaacctgaaataaaactggctcagaatgtaacaggaaattctggcata aatttgacctgcacgtctaagcaaggtcacccgaaacctaagaagatgtattttctgataactaattcaac taatgagtatggtgataacatgcagatatcacaagataatgtcacagaactgttcagtatctccaacagc ctctctctttcattcccggatggtgtgtggcatatgaccgttgtgtgtgttctggaaacggagtcaatgaagat ttcctccaaacctctcaatttcactcaagagtttccatctcctcaaacgtattggaaggagattacagcttca gttactgtggccctcctccttgtgatgctgctcatcattgtatgtcacaagaagccgaatcagcctagcagg cccagcaacacagcctctaagttagagcgggatagtaacgctgacagagagactatcaacctgaag gaacttgaaccccaaattgcttcagcaaaaccaaatgcagagtgtactagtCACTATCCCATA GTCccactggacagcaagatctcgctgcccatccggaatggctctgaagtgcgggacccactggtca cctatgagggatctctcccaccagcatccccgctgcaagacaacctggttatcgccctgcacagttatga gccctcccatgatggagacttgggctttgagaagggtgaacagctccgaatcctggagcagagcggtg agtggtggaaggctcagtccctgacgactggccaagaaggcttcattcccttcaacttcgtggcgaaag caaacagcctggagcctgaaccttggttcttcaagaatctgagccgtaaggacgccgagcggcagcttt tggcgcccgggaacacgcatggatccttcctgatccgggaaagcgaaagcactgcggggtccttttcc ctgtcggtcagagacttcgaccagaaccagggagaagtggtgaaacattacaagatccgtaacctag acaacggtggcttctacatctcccctcgtatcacttttcccggattgcacgatctagtccgccattacacca acgcctctgatgggctgtgcacaaagttgagccgtccttgccagacccagaagccccagaaaccatg gtgggaggacgaatgggaagttcccagggaaacactgaagttggtggagcggctgggagctggcca gttcggggaagtgtggatggggtactacaacggacacacgaaggtggcggtgaagagtctgaaaca agggagcatgtcccccgacgccttcctggctgaggctaacctcatgaagcagctgcagcacccgcgg ctagtccggctttatgcagtggtcacccaggaacccatctacatcatcacggaatacatggagaacggg agcctagtagattttctcaagactccctcgggcatcaagttgaatgtcaacaaacttttggacatggcagc ccagattgcagagggcatggcgttcatcgaagaacagaattacatccatcgggacctgcgccgccgcc aacatcctggtgtctgacacgctgagctgcaagattgcagactttggcctggcgcgcctcattgaggaca atgagtacacggcccgggaggggccaaatttcccattaagtggacagcaccagaagccattaacta tgggaccttcaccatcaagtcagacgtgtggtccttcgggatcttgcttacagagatcgtcacccacggtc gaatccctacccaggaatgaccaaccctgaagtcattcagaacctggagagaggctaccgcatggtg agacctgacaactgtccggaagagctgtaccacctcatgatgctgtgctggaaggagcgcccagagg accggcccacgtttgactaccttcggagtgttctggatgacttcttcacagccacagagggcCAGTAC CAGCCCCAGCCTggtacctagtgagaattctacatg

[0077] The disclosures of the following U.S. Patents are incorporated in their entirety by reference herein: U.S. Pat. Application No. 20140219975; U.S. Pat. No. 8450112; U.S.

Pat. No. 7741465; U.S. Pat. No. 6319494; CA 2209300; CA 2104957; EP 0574512; U.S. Pat. No. 6407221; U.S. Pat. No. 6268411; U.S. Pat. Application No. 20040258697; EP 1292621; EP 2659893; WO 2011101681; WO 2005054292; EP 1379670; U.S. Pat. No. 6056952; U.S. Pat. No. 6410319; U.S. Pat. No. 8524234; U.S. Pat. No. 7871817.

[0078] As used herein, the term "about" refers to plus or minus 10% of the referenced number.

[0079] Various modifications of the invention, in addition to those described herein, will be apparent to those skilled in the art from the foregoing description. Such modifications are also intended to fall within the scope of the appended claims. Each reference cited in the present application is incorporated herein by reference in its entirety.

[0080] Although there has been shown and described the preferred embodiment of the present invention, it will be readily apparent to those skilled in the art that modifications may be made thereto which do not exceed the scope of the appended claims. Therefore, the scope of the invention is only to be limited by the following claims. In some embodiments, the figures presented in this patent application are drawn to scale, including the angles, ratios of dimensions, etc. In some embodiments, the figures are representative only and the claims are not limited by the dimensions of the figures. In some embodiments, descriptions of the inventions described herein using the phrase "comprising" includes embodiments that could be described as "consisting of", and as such the written description requirement for claiming one or more embodiments of the present invention using the phrase "consisting of" is met.

WHAT IS CLAIMED IS:

1. An engineered cell co-expressing on its surface:
   a. a chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion derived from a MHC protein directly or indirectly fused to a T cell receptor (TCR) portion derived from a TCR protein; and
   b. a surrogate coreceptor (SCR) comprising a cell surface receptor ligand portion directly or indirectly fused to a signaling molecule portion;
   wherein the MHCR is adapted to bind to a TCR of a target cell and the SCR is adapted to bind to a cell surface receptor of the target cell.
2. The engineered cell of claim 1, wherein binding of the MHCR to the TCR of the target cell and binding of the SCR to the cell surface receptor of the target cell (i) initiates a signaling cascade effective for eliminating the target cell or (ii) instructs the target cell to differentiate to a specific effector function.
3. The engineered cell of claim 1, wherein the cell is a T cell.
4. The engineered cell of claim 1, wherein the MHC portion of the MHCR is N-terminal to the TCR portion of the MHCR.
5. The engineered cell of claim 1, wherein the MHC portion is indirectly fused to the TCR portion via a linker.
6. The engineered cell of claim 5, wherein the linker comprises a glycine-rich peptide.
7. The engineered cell of claim 1, wherein the MHC protein, the TCR protein, or both the MHC protein and the TCR protein are mammalian proteins.
8. The engineered cell of claim 7, wherein the mammal is a human or a mouse.
9. The engineered cell of claim 1, wherein the TCR portion comprises at least a portion of a transmembrane domain of the TCR protein and the MHC portion comprises at least a portion of an extracellular domain of the MHC protein.
10. The engineered cell of claim 1, wherein the TCR portion comprises at least a portion of a transmembrane domain and at least a portion of a cytoplasmic domain of a TCR protein, and the MHC portion comprises at least a portion of an extracellular domain of the MHC protein.
11. The engineered cell of claim 1, wherein the MHCR further comprises a peptide antigen integrated into the MHC portion, or directly or indirectly fused to the MHC portion.

12. The engineered cell of claim 1, wherein the signaling molecule portion has kinase or phosphatase activity.
13. The engineered cell of claim 1, wherein the signaling molecule portion comprises a Src kinase.
14. The engineered cell of claim 1, wherein the MHC protein comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a fragment thereof, or a combination thereof.
15. The engineered cell of claim 1, wherein the MHC molecule comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a peptide that is at least 90% identical to HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, or H2-EK beta, a fragment thereof, or a combination thereof.
16. The engineered cell of claim 1, wherein the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a fragment thereof, or a combination thereof.
17. The engineered cell of claim 1, wherein the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a peptide that is at least 90% identical to TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, or TCC4, a fragment thereof, or a combination thereof.
18. The engineered cell of claim 1, wherein the cell surface receptor ligand portion of the SCR comprises a CD28 ligand, a CTLA-4 ligand, an ICOS ligand, an OX4O ligand, a PD-1 ligand, or a CD2 ligand.
19. The engineered cell of claim 18, wherein the CD28 ligand comprises CD80, CD86, or both CD80 and CD86.
20. The engineered cell of claim 1, wherein the MHCR is adapted to complex with a CD3 subunit.
21. The engineered cell of claim 1 further co-expressing a second SCR.
22. A chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion derived from a MHC protein directly or indirectly fused to a T cell

receptor (TCR) portion derived from a TCR protein, wherein the MHCR is adapted to bind to a TCR of a target cell.

23. The MHCR of claim 22, wherein binding of the MHCR to the TCR of the target cell (i) initiates a signaling cascade effective for eliminating the target cell or (ii) instructs the target cell to differentiate to a specific effector function.
24. The MHCR of claim 22, wherein the cell is a T cell.
25. The MHCR of claim 22, wherein the MHC portion of the MHCR is N-terminal to the TCR portion of the MHCR.
26. The MHCR of claim 22, wherein the MHC portion is indirectly fused to the TCR portion via a linker.
27. The MHCR of claim 26, wherein the linker comprises a glycine-rich peptide.
28. The MHCR of claim 22, wherein the MHC protein, the TCR protein, or both the MHC protein and the TCR protein are mammalian proteins.
29. The MHCR of claim 28, wherein the mammal is a human or a mouse.
30. The MHCR of claim 22, wherein the TCR portion comprises at least a portion of a transmembrane domain and at least a portion of a cytoplasmic domain of a TCR protein, and the MHC portion comprises at least a portion of an extracellular domain of the MHC protein.
31. The MHCR of claim 22, wherein the MHCR further comprises a peptide antigen integrated into the MHC portion, or directly or indirectly fused to the MHC portion.
32. The MHCR of claim 22, wherein the MHC protein comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a fragment thereof, or a combination thereof.
33. The MHCR of claim 22, wherein the MHC molecule comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a peptide that is at least 90% identical to HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, or H2-EK beta, a fragment thereof, or a combination thereof.
34. The MHCR of claim 22, wherein the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a fragment thereof, or a combination thereof.

35. The MHCR of claim 22, wherein the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a peptide that is at least 90% identical to TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, or TCC4, a fragment thereof, or a combination thereof.
36. The MHCR of claim 22, wherein the MHCR is adapted to complex with a CD3 subunit.
37. A method of eliminating a target cell or reprogramming a target cell, said target cell comprising a TCR, said method comprising introducing a genetically engineered cell that expresses on its surface a chimeric receptor (MHCR) according to any of claims 1-21 to the target cell, the MHCR is specific for the TCR of the target cell, wherein upon binding of the MHCR to the TCR the genetically engineered cell (a) initiates a signaling cascade that eliminates the target cell, or (b) instructs the target cell to differentiate to a specific effector function.
38. The method of claim 37, wherein the method is for immunotherapy.
39. The method of claim 37, wherein the target cell is an autoreactive T cell.
40. A vector encoding the MHCR of any of Claims 1-21.
41. A vector encoding the SCR of any of Claims 1-21.
42. An engineered cell co-expressing on its surface:
    a. a chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion derived from an extracellular domain of a mammalian MHC protein directly or indirectly linked to a transmembrane domain of a T cell receptor (TCR) portion derived from a mammalian TCR protein, the MHC portion being N-terminal to the TCR portion; and
    b. a surrogate coreceptor (SCR) comprising a cell surface receptor ligand portion indirectly linked to a signaling molecule portion by a transmembrane domain, the signaling molecule portion having kinase or phosphatase activity;

    wherein the MHCR is adapted to bind to a TCR of a target cell and the SCR is adapted to bind to a cell surface receptor of the target cell.
43. The engineered cell of claim 42, wherein the cell is a T cell.
44. The engineered cell of claim 42, wherein the MHCR further comprises a peptide

antigen integrated into the MHC portion, or directly or indirectly fused to the MHC portion.

45. The engineered cell of claim 42, wherein the signaling molecule portion comprises a Src kinase.

46. The engineered cell of claim 42, wherein the MHC protein comprises HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, H2-EK beta, a fragment thereof, or a combination thereof.

47. The engineered cell of claim 42, wherein the TCR molecule comprises TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4, a fragment thereof, or a combination thereof.

48. An engineered T-cell co-expressing on its surface:

a. a chimeric receptor (MHCR) comprising a major histocompatibility complex (MHC) portion derived from an extracellular domain of a mammalian MHC protein directly or indirectly linked to a transmembrane domain of a T cell receptor (TCR) portion derived from a mammalian TCR protein, the MHC portion being N-terminal to the TCR portion, the MHC portion being selected from HLA-A, HLA-B, HLA-C, HLA-DPA1, HLA-DPB1, HLA-DQA1, HLA-DQB1, HLA-DRA, HLA-DRB, H2-Aa, H2-B1, H2-K1, H2-EB beta, H2-EK alpha, and H2-EK beta, the TCR portion being selected from TRAC, TRBC1, TRBC2, TRDC, TRGC1, TRGC2, TCRA, TCB1, TCB2, TCC1, TCC2, TCC3, TCC4; and

b. a surrogate coreceptor (SCR) comprising a cell surface receptor ligand portion indirectly linked to a signaling molecule portion by a transmembrane domain, the signaling molecule portion having kinase or phosphatase activity;

wherein the MHCR is adapted to bind to a TCR of a target cell and the SCR is adapted to bind to a cell surface receptor of the target cell.

49. The engineered T-cell of claim 48, wherein the MHCR further comprises a peptide antigen integrated into the MHC portion, or directly or indirectly fused to the MHC portion.

50. A surrogate coreceptor (SCR) comprising a cell surface receptor ligand portion directly or indirectly fused to a signaling molecule portion via a transmembrane

domain, wherein the SCR is adapted to bind to a cell surface receptor of a target cell.

51. The SCR of claim 50, wherein the cell surface receptor ligand portion is indirectly fused to the signaling molecule portion via a linker.

52. The SCR of claim 50, wherein the signaling molecule portion has kinase or phosphatase activity.

53. The SCR of claim 50, wherein the signaling molecule portion comprises a Src kinase.

54. The SCR of claim 50, wherein the cell surface receptor ligand portion of the SCR comprises a CD28 ligand, a CTLA-4 ligand, an ICOS ligand, an OX4O ligand, a PD-1 ligand, or a CD2 ligand.

55. The SCR of claim 54, wherein the CD28 ligand comprises CD80, CD86, or both CD80 and CD86.

FIG. 1A

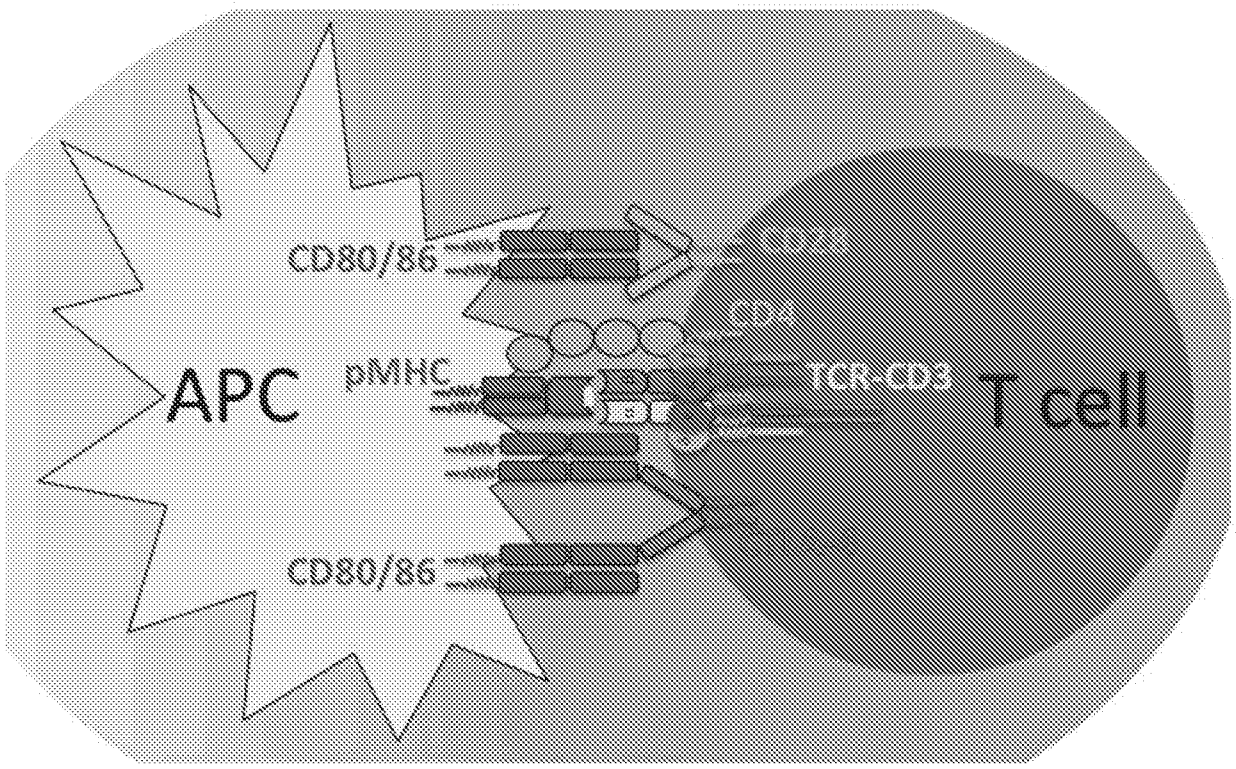

CD80/86
APC
pMHC
TCR-CD3
T cell
CD80/86

FIG. 1B

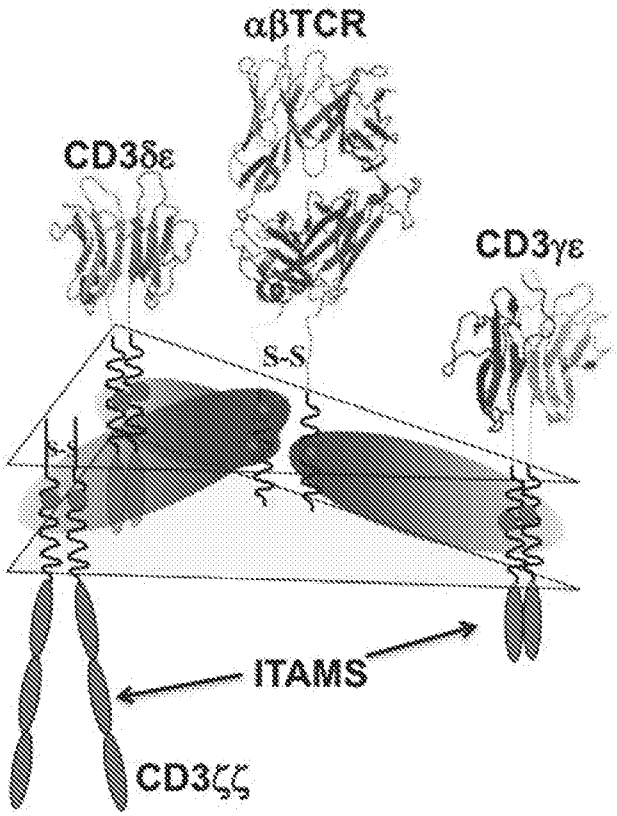

αβTCR
CD3δε
CD3γε
S-S
ITAMS
CD3ζζ

FIG. 2A

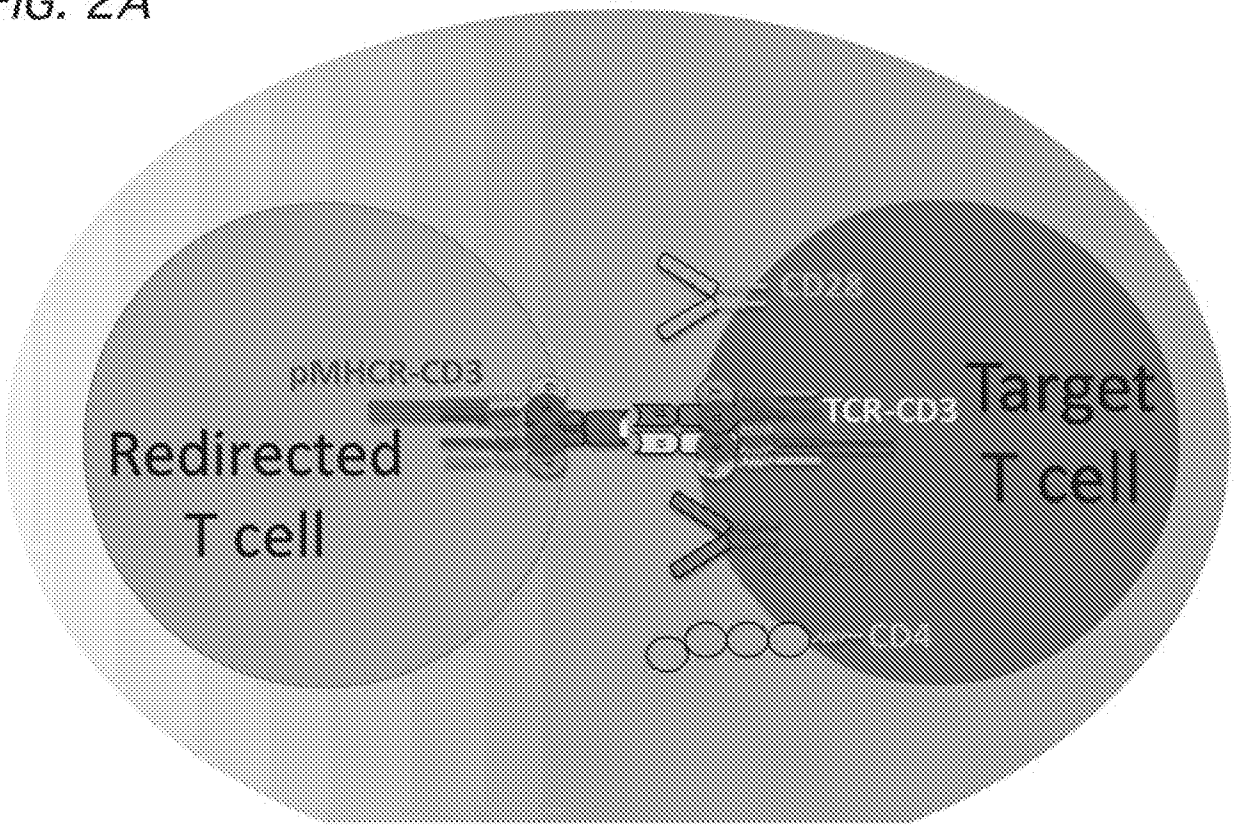

pMHCR-CD3
Redirected
T cell
TCR-CD3
Target
T cell

FIG. 2B

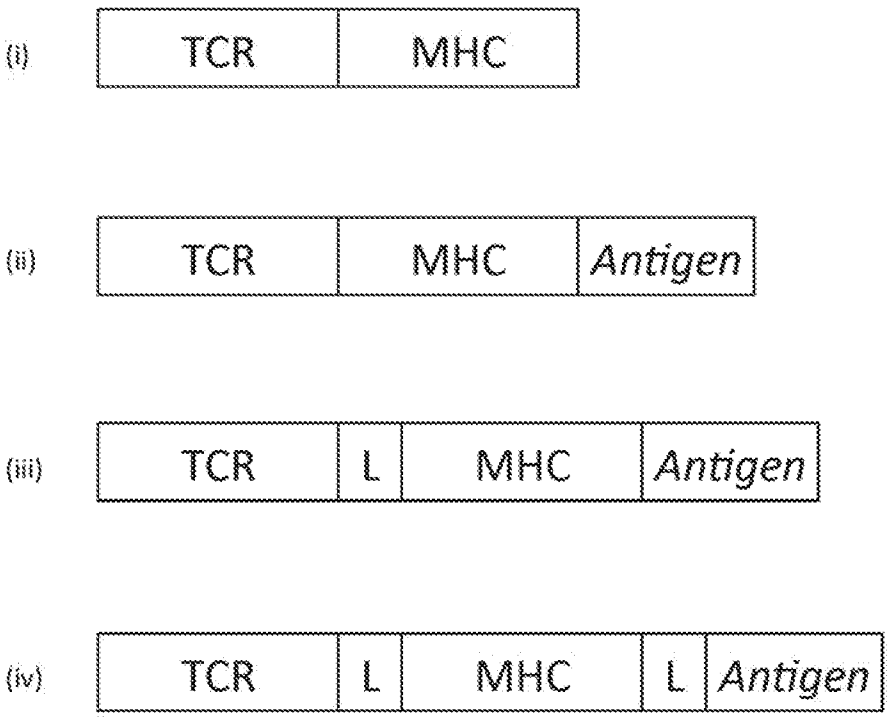

(i)
TCR
MHC
(ii)
TCR
MHC
Antigen
(iii)
TCR
L
MHC
Antigen
(iv)
TCR
L
MHC
L
Antigen

FIG.3A

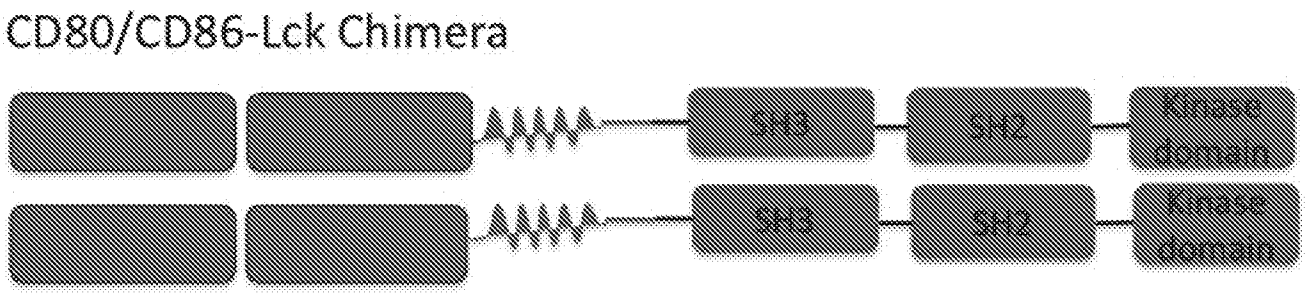

CD80/CD86-Lck Chimera
SH3
SH2
Kinase domain
SH3
SH2
Kinase domain

FIG.3B

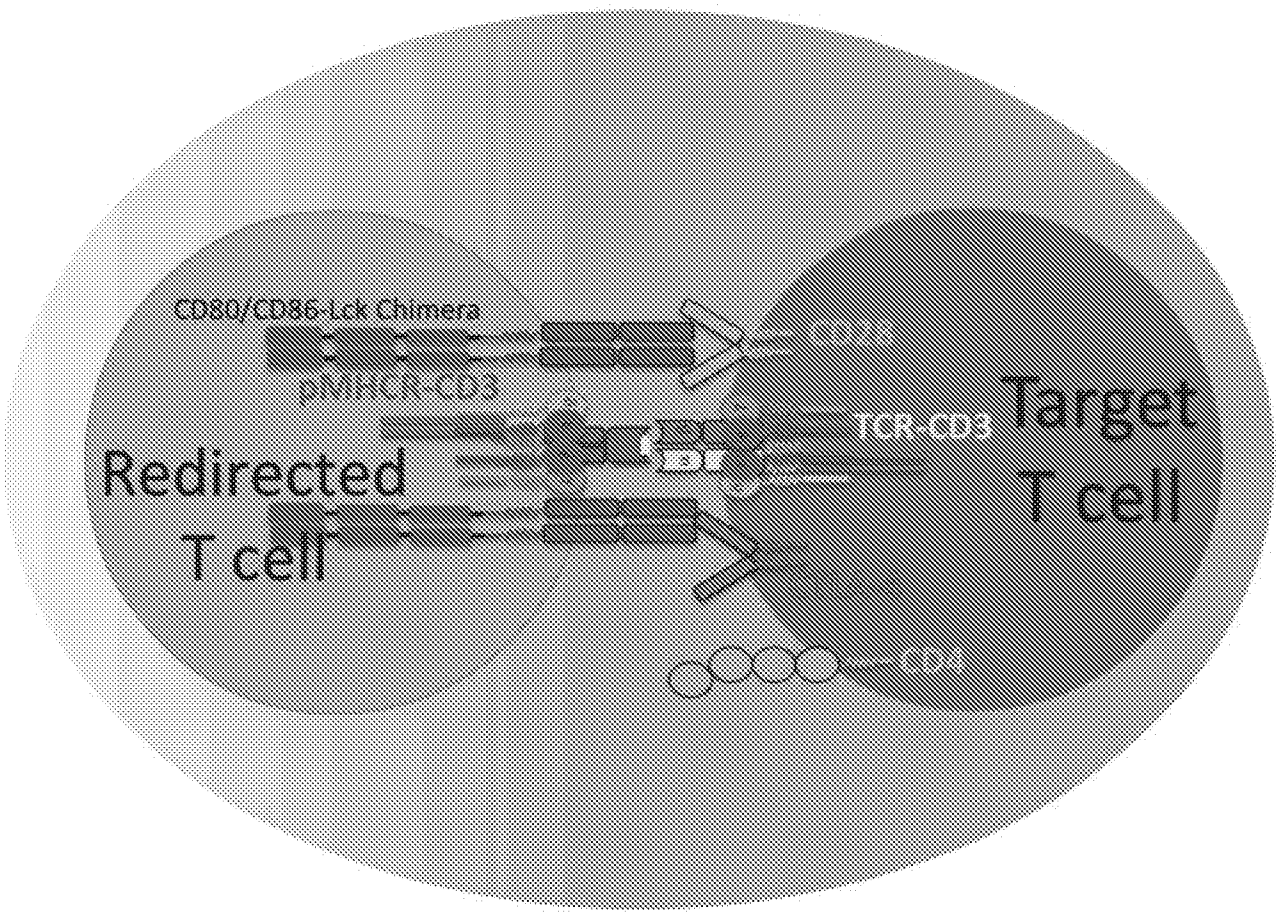

CD80/CD86-Lck Chimera
pMHC
TCR-CD3
Redirected T cell
TCR-CD3
Target T cell
CD19

FIG. 4

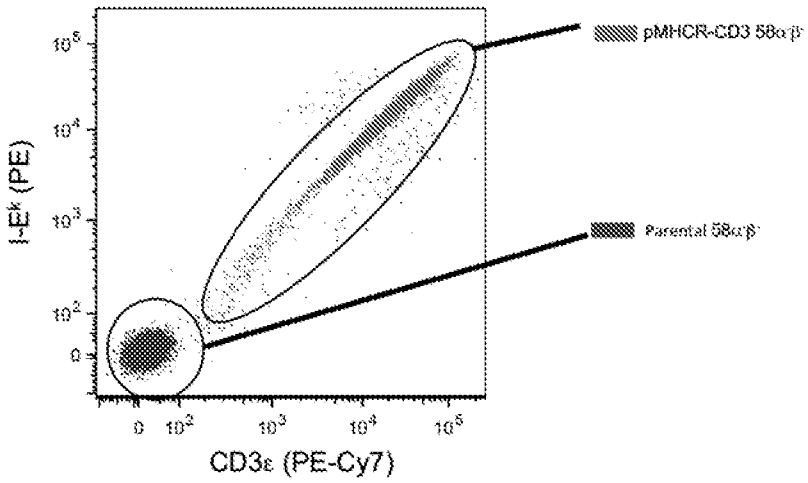


FIG. 5

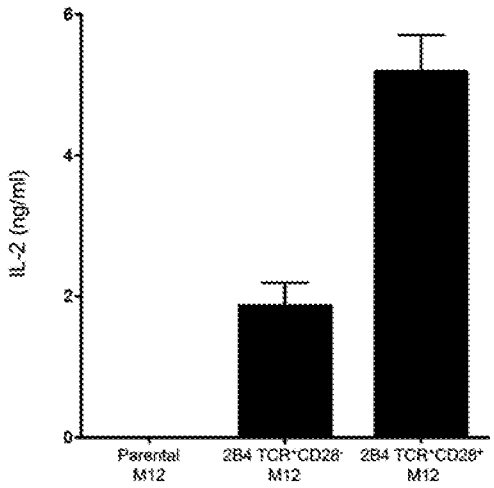
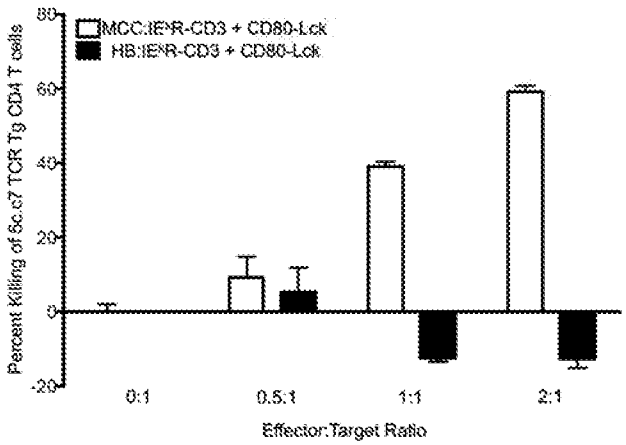


FIG. 6



INTERNATIONAL SEARCH REPORT

International application No.
PCT/US16/40177

A. CLASSIFICATION OF SUBJECT MATTER
IPC(8) - A61K 38/17; C07K 14/705, 14/725; C12N 5/10 (2016.01)
CPC - A61K 38/1774; C07K 14/70503, 14/70532, 14/70521, 14/7051, 14/70539, 14/70507; C12N 5/10
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC(8) Classifications: A61K 38/00, 38/17; G01N 33/50, 33/68; C07K 7/06, 14/62, 14/705, 14/725; C12N 5/10, 5/12 (2016.01)
CPC Classifications: A61K 38/00, 38/17, 38/1774; G01N 33/50, 33/68, 33/6893; C07K 7/06, 14/62, 14/705, 14/70503, 14/70532

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)
PatSeer (US, EP, WO, JP, DE, GB, CN, FR, KR, ES, AU, IN, CA, INPADOC Data); Google; Google Scholar; PubMed; EBSCO; Engineer*, Recombinant*, redirect*, cell*, express*, Chimer*, receptor*, MHCR, MHC, 'Major histocompatibility complex', 'Human leukocyte antigen', HLA, TCR, 'T cell receptor', Surrogat*, substitut*, prox*, replacement*, coreceptor*, SCR, surfac*, ligand*, Cd28,

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US 2014/0219975 A1 (JUNE, CH et al.) 07 August 2014; paragraphs [0008], [0032], [0089], [0164]; [0184]; claims 3, 4.	1-21, 37/1-21, 38/37/1-21, 39/37/1-21, 40/1-21, 41/1-21, 42- 55
A	(QIAN, Z et al.) Engineered Tregulatory Cells Co-expressing MHC Class II:peptide Complexes Are Efficient Inhibitors Of Autoimmune T Cell Function And Prevent The Development Of Autoimmune Arthritis. J. Immunol. Author manuscript. 1 June 2014, Vol. 190; pages 1-23; abstract; page 3, fifth paragraph-page 4, first paragraph; page 6, fourth paragraph; doi:10.4049/jimmunol.1300024.	1-36, 37/1-21, 38/37/1-21, 39/37/1-21, 40/1-21, 41/1-21, 42-49
A	(THIEL, M et al.) Efficiency Of T-cell Costimulation By CD80 and CD86 Cross-linking Correlates With Calcium Entry. Immunology. 2009, Vol. 129, pages 28-40; page 28, first column, first paragraph-second column, first paragraph; page 28, second column, second paragraph; page 29, first column, third paragraph; page 32, first column, first paragraph; doi:10.1111/j.1365-2567.2009.03155.x.	1-21, 37/1-21, 38/37/1-21, 39/37/1-21, 40/1-21, 41/1-21, 42-55
A	US 8,906,383 B2 (PEAKMAN, M et al.) 09 December 2014; column 4, line 66- column 5, line 1; column 8, line 21.	1-36, 37/1-21, 38/37/1-21, 39/37/1-21, 40/1-21, 41/1-21, 42-49
A	(KUHNS, MS et al.) TCR Signaling Emerges From The Sum Of Many Parts. Frontiers In Immunology. 25 June 2012, Vol. 3, pages 1-13; figure 2; page 5, second column, second paragraph; page 6, first column, second paragraph; doi: 10.3389/fimmu.2012.00159.	22-36
A	WO 2014/117121 A1 (ST. JUDE CHILDREN'S RESEARCH HOSPITAL, INC. et al.) 31 July 2014; abstract; paragraphs [0005], [00195].	22-36

☒ Further documents are listed in the continuation of Box C. ☐ See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" earlier application or patent but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed
"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
12 September 2016 (12.09.2016)	30 SEP 2016
Name and mailing address of the ISA/ Mail Stop PCT, Attn: ISA/US, Commissioner for Patents P.O. Box 1450, Alexandria, Virginia 22313-1450 Facsimile No. 571-273-8300	Authorized officer Shane Thomas PCT Helpdesk: 571-272-4300 PCT OSP: 571-272-7774

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US16/40177

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	(BUENO, C et al.) T Cell Signalling Induced By Bacterial Superantigens. Chemical Immunology And Allergy. February 2007, Vol. 93, pages 161-180, DOI: 10.1159/0000100894; page 171, third paragraph; page 172, first paragraph.	50-55
A	(PODOJIL, JR et al.) Molecular Mechanisms Of T Cell Receptor And Costimulatory Molecule Ligation/Blockade In Autoimmune Disease Therapy. Immunol Rev. Author manuscript. 1 May 2010, Vol. 229, pages 1-28, doi:10.1111/j.1600-065X.2009.00773.x; page 2, first paragraph; page 11, second paragraph.	50-55