

CLAIMS

What is claimed is:

1. A chimeric antigen receptor comprising: an ectodomain comprising at least one antigen recognition domain; a transmembrane domain; and an endodomain comprising at least one signaling domain;

wherein the at least one signaling domain is originated from a cytoplasmic domain of a signal transducing protein specific to T and/or NK cell activation or functioning;

wherein the chimeric antigen receptor, when comprised in an induced pluripotent stem cell (iPSC), promotes differentiation of the iPSC directed to a desired derivative effector cell, and wherein the iPSC-derived effector cell differentiated from the iPSC has at least one of the following characteristics comprising:

- (i) improved persistency and/or survival;
- (ii) improved cell expansion;
- (iii) increased cytotoxicity;
- (iv) increased resistance to allojection;
- (v) improved tumor penetration;
- (vi) enhanced ability in migrating, and/or activating or recruiting bystander immune cells, to tumor sites; and
- (vii) enhanced ability to reduce tumor immunosuppression,

in comparison to a primary immune cell obtained from peripheral blood, umbilical cord blood, or any other donor tissues.

2. The chimeric antigen receptor of claim 1, wherein

(a) the signal transducing protein comprises any one of: 2B4 (Natural killer Cell Receptor 2B4), 4-1BB (Tumor necrosis factor receptor superfamily member 9), CD16 (IgG Fc region Receptor III/A), CD2 (T-cell surface antigen CD2), CD28 (T-cell-specific surface glycoprotein CD28), CD28H (Transmembrane and immunoglobulin domain-containing protein 2), CD3 ζ (T-cell surface glycoprotein CD3 zeta chain), DAP10 (Hematopoietic cell signal transducer), DAP12 (TYRO protein tyrosine kinase-binding protein), DNAM1 (CD226 antigen), Fc ϵ RI γ (High affinity immunoglobulin epsilon receptor subunit gamma), IL21R (Interleukin-21 receptor), IL-2R β /IL-15RB (Interleukin-2 receptor subunit beta), IL-2R γ (Cytokine receptor common subunit gamma), IL-7R (Interleukin-7 receptor subunit alpha), KIR2DS2 (Killer cell immunoglobulin-like receptor 2DS2), NKG2D (NKG2-D type II integral membrane protein), NKp30 (Natural cytotoxicity triggering receptor 3), NKp44 (Natural cytotoxicity triggering

receptor 2), NKp46 (Natural cytotoxicity triggering receptor 1), CS1(SLAM family member 7), and CD8 (T-cell surface glycoprotein CD8 alpha chain); and/or

(b) the at least one signaling domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of 2B4, 4-1BB, CD16, CD2, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ IL21R, IL-2R β (IL-15R β), IL-2R γ , IL-7R, KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CD3 ζ 1XX, CS1, or CD8, represented by SEQ ID NOs: 21-41, 54 and 56, respectively.

3. The chimeric antigen receptor of claim 2, wherein the at least one signaling domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of 2B4, 4-1BB, CD16, CD2, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ IL21R, IL-2R β (IL-15R β), IL-2R γ , IL-7R, KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CD3 ζ 1XX, CS1, or CD8, represented by SEQ ID NOs: 21-41, 54 and 56, respectively; and wherein the portion of said cytoplasmic domain comprises an ITAM (immunoreceptor tyrosine-based activation motif), a YxxM motif, a TxYxxV/I motif, FcR γ , a hemi-ITAM, and/or an ITT-like motif.

4. The chimeric antigen receptor of claim 1, wherein the endodomain comprises a first signaling domain, a second signaling domain, and optionally a third signaling domain; and wherein the first, second and third signaling domains are different.

5. The chimeric antigen receptor of claim 4, wherein the second or the third signaling domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of 2B4, 4-1BB, CD16, CD2, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ IL21R, IL-2R β (IL-15R β), IL-2R γ , IL-7R, KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CD3 ζ 1XX, CS1, or CD8, represented by SEQ ID NOs: 21-41, 54 and 56, respectively.

6. The chimeric antigen receptor of claim 1, wherein the endodomain comprises only one signaling domain, wherein the endodomain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of DNAM1, CD28H, KIR2DS2, DAP12 or DAP10.

7. The chimeric antigen receptor of claim 4, wherein the endodomain comprises two different signaling domains, and wherein said endodomain domain comprises fused cytoplasmic domains, or portions thereof, in any one of the forms: 2B4-CD3 ζ /1XX, 2B4-DNAM1, 2B4-FcERI γ , 2B4-DAP10, CD16-DNAM1, CD16-DAP10, CD16-DAP12, CD2-CD3 ζ /1XX, CD2-DNAM1, CD2-FcERI γ , CD2-DAP10, CD28-DNAM1, CD28-FcERI γ , CD28-DAP10, CD28-DAP12, CD28H-CD3 ζ /1XX, DAP10-CD3 ζ /1XX, DAP10-DAP12, DAP12-CD3 ζ /1XX, DAP12-DAP10, DNAM1-CD3 ζ /1XX, KIR2DS2-CD3 ζ /1XX, KIR2DS2-DAP10, KIR2DS2-2B4, and NKp46-2B4.
8. The chimeric antigen receptor of claim 4, wherein the endodomain comprises three different signaling domains, and wherein said endodomain domain comprises fused cytoplasmic domains, or portions thereof, in any one of the forms comprising 2B4-DAP10-CD3 ζ /1XX, 2B4-IL21R-DAP10, 2B4-IL2RB-DAP10, 2B4-IL2RB-CD3 ζ /1XX, 2B4-41BB-DAP10, CD16-2B4-DAP10, and KIR2DS2-2B4-CD3 ζ /1XX.
9. The chimeric antigen receptor of claim 1, wherein the transmembrane domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to a transmembrane region, or a portion thereof, of CD2, CD3D, CD3E, CD3G, CD3 ζ , CD4, CD8, CD8a, CD8b, CD16, CD27, CD28, CD28H, CD40, CD84, CD166, 4-1BB, OX40, ICOS, ICAM-1, CTLA4, PD1, LAG3, 2B4, BTLA, DNAM1, DAP10, DAP12, FcERI γ , IL7, IL12, IL15, KIR2DL4, KIR2DS1, KIR2DS2, NKp30, NKp44, NKp46, NKG2C, NKG2D, CS1, or T cell receptor polypeptide.
10. The chimeric antigen receptor of claim 1, wherein the transmembrane domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to a transmembrane region, or a portion thereof, of 2B4, CD2, CD16, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ , KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CS1, or CD8, represented by SEQ ID NOs: 1-20, 53 and 55, respectively.
11. The chimeric antigen receptor of claim 1, wherein the transmembrane domain and its immediately linked signaling domain are from a same protein or from different proteins.
12. The chimeric antigen receptor of claim 1, wherein the chimeric antigen receptor comprises a transmembrane domain and an endodomain (*TM*-(endodomain)), and wherein the chimeric antigen receptor comprises:

- (i) one of the forms: *NKG2D*-(2B4-IL2RB-CD3 ζ), *CD8*-(41BB- CD3 ζ 1XX), *CD28*-(CD28-2B4-CD3 ζ), *CD28H*-(CD28H-CD3 ζ), *DNAM1*-(DNAM1-CD3 ζ), *DAP10*-(DAP10-CD3 ζ), *KIR2DS2*-(KIR2DS2-CD3 ζ), *KIR2DS2*-(KIR2DS2-DAP10), *KIR2DS2*-(KIR2DS2-2B4), *CD16*-(CD16-2B4-DAP10), *CD16*-(CD16-DNAM1), *NKp46*-(NKp46-2B4), *NKp46*-(NKp46-2B4-CD3 ζ), *NKp46*-(NKp46-CD2-Dap10), *CD2*-(CD2-CD3 ζ), *2B4*-(2B4-CD3 ζ), *2B4*-(2B4-Fc ϵ RIg), and *CS1*-(CS1-CD3 ζ); or
- (ii) an amino acid sequence having about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, about 99%, or 100% identity to a sequence represent by each of SEQ ID NOs: 57-74.

13. The chimeric antigen receptor of claim 1, wherein the antigen recognition domain specifically binds an antigen associated with a disease, a pathogen, a liquid tumor, or a solid tumor.

14. The chimeric antigen receptor of claim 1, wherein the antigen recognition domain is specific to:

- (i) any one of CD19, BCMA, CD20, CD22, CD38, CD123, HER2, CD52, EGFR, GD2, MICA/B, MSLN, VEGF-R2, PSMA and PDL1; or
- (ii) any one of ADGRE2, carbonic anhydrase IX (CAIX), CCRI, CCR4, carcinoembryonic antigen (CEA), CD3, CD5, CD7, CD8, CD10, CD20, CD22, CD30, CD33, CD34, CD38, CD41, CD44, CD44V6, CD49f, CD56, CD70, CD74, CD99, CD123, CD133, CD138, CDS, CLEC12A, an antigen of a cytomegalovirus (CMV) infected cell, epithelial glycoprotein2 (EGP 2), epithelial glycoprotein-40 (EGP-40), epithelial cell adhesion molecule (EpCAM), EGFR γ III, receptor tyrosine-protein kinases erb- B2,3,4, EGFR, EGFR-VIII, ERBB folate-binding protein (FBP), fetal acetylcholine receptor (AChR), folate receptor-a, Ganglioside G2 (GD2), Ganglioside G3 (GD3), human Epidermal Growth Factor Receptor 2 (HER-2), human telomerase reverse transcriptase (hTERT), ICAM-1, Integrin B7, Interleukin-13 receptor subunit alpha-2 (IL-13R α 2), κ -light chain, kinase insert domain receptor (KDR), Lewis A (CA19.9), Lewis Y (LeY), L1 cell adhesion molecule (L1-CAM), LILRB2, melanoma antigen family A 1 (MAGE-A1), MICA/B, Mucin 1 (Muc-1), Mucin 16 (Muc-16), Mesothelin (MSLN), NKCSI, NKG2D ligands, c-Met, cancer-testis antigen

NY-ESO-1, oncofetal antigen (h5T4), PRAME, prostate stem cell antigen (PSCA), PRAME prostate-specific membrane antigen (PSMA), tumor-associated glycoprotein 72 (TAG-72), TIM-3, TRBC1, TRBC2, vascular endothelial growth factor R2 (VEGF- R2), Wilms tumor protein (WT-1), and a pathogen antigen.

15. The chimeric antigen receptor of claim 1, wherein the ectodomain comprises one or more of:

- (i) two antigen recognition domains;
- (ii) a signal peptide; and/or
- (iii) a spacer/hinge.

16. The chimeric antigen receptor of claim 1, wherein the chimeric antigen receptor is comprised in a bi-cistronic construct co-expressing a partial or full length peptide of a cell surface expressed exogenous cytokine or a receptor thereof, wherein the exogenous cytokine or receptor thereof comprises:

- (a) at least one of IL2, IL4, IL6, IL7, IL9, IL10, IL11, IL12, IL15, IL18, IL21, and its respective receptor(s); or
- (b) at least one of:
 - (i) co-expression of IL15 and IL15R α by using a self-cleaving peptide;
 - (ii) a fusion protein of IL15 and IL15R α ;
 - (iii) an IL15/IL15R α fusion protein with intracellular domain of IL15R α truncated or eliminated;
 - (iv) a fusion protein of IL15 and membrane bound Sushi domain of IL15R α ;
 - (v) a fusion protein of IL15 and IL15R β ;
 - (vi) a fusion protein of IL15 and common receptor γ C, wherein the common receptor γ C is native or modified; and
 - (vii) a homodimer of IL15R β .

17. The chimeric antigen receptor of claim 1, wherein the derivative effector cell from iPSC differentiation comprises one or more of: a derivative CD34 cell, a derivative hematopoietic stem and progenitor cell, a derivative hematopoietic multipotent progenitor cell, a derivative T cell progenitor, a derivative NK cell progenitor, a derivative T cell, a derivative NKT cell, a derivative NK cell, a derivative B cell, or a derivative immune effector cell.

18. The chimeric antigen receptor of claim 17, wherein the iPSC-derived immune effector cell expresses said chimeric antigen receptor, and comprises at least one functional feature that is not present in a primary T, NK, and/or NKT cell.
19. A cell or a population thereof, wherein:
- (i) the cell is an immune cell, an induced pluripotent cell (iPSC), a clonal iPSC, or an iPS cell line cell; or the cell is a derivative effector cell obtained from differentiating the iPSC; and
 - (ii) the cell comprises at least one chimeric antigen receptor (CAR) of any one of claims 1-18.
20. The cell or population thereof of claim 19, wherein the derivative effector cell is a hematopoietic cell, and comprises longer telomeres in comparison to its counterpart primary cell obtained from peripheral blood, umbilical cord blood, or any other donor tissues; or wherein the CAR has at least one of the following characteristics:
- (i) being T or NK cell specific;
 - (ii) bi-specific in antigen binding;
 - (iii) being a switchable CAR;
 - (iv) being a dimerized CAR;
 - (v) being a split CAR;
 - (vi) being a multi-chain CAR;
 - (vii) being an inducible CAR; and
 - (viii) being inserted at one of the following gene loci: B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR α or β constant region, NKG2A, NKG2D, CD38, CD25, CD69, CD44, CD58, CD54, CD56, CIS, CBLB, SOCS2, PD1, CTLA4, LAG3, TIM3, or TIGIT, wherein the insertion knocks out or reduces expression of the gene in the locus.
21. The cell or population thereof of claim 19, wherein the cell further comprises one or more of:
- (i) CD38 knockout;
 - (ii) B2M null or low, and optionally CIITA null or low, in comparison to its counterpart primary cell;
 - (iii) introduced expression of HLA-G or non-cleavable HLA-G, or knockout of one or both of CD58 and CD54;

- (iv) a CD16 or a variant thereof;
- (v) a second CAR having different targeting specificity;
- (vi) a partial or full peptide of a cell surface expressed exogenous cytokine and/or a receptor thereof;
- (vii) at least one of the genotypes listed in Table 2;
- (viii) deletion or reduced expression in at least one of TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR, NKG2A, NKG2D, CD25, CD69, CD44, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, and TIGIT, in comparison to its counterpart primary cell; or
- (ix) introduced or increased expression in at least one of HLA-E, 41BBL, CD3, CD4, CD8, CD16, CD47, CD113, CD131, CD137, CD80, PDL1, A_{2A}R, antigen-specific TCR, Fc receptor, an engager, and surface triggering receptor for coupling with an agonist, in comparison to its counterpart primary cell.

22. The cell or population thereof of claim 19, wherein the cell is a derivative effector cell, and has at least one of the following characteristics comprising:

- (i) improved persistency and/or survival;
- (ii) increased resistance to alloreactive recipient immune cells;
- (iii) increased cytotoxicity;
- (iv) improved tumor penetration;
- (v) enhanced or acquired ADCC;
- (vi) enhanced ability in migrating, and/or activating or recruiting bystander immune cells, to tumor sites;
- (vii) enhanced ability to reduce tumor immunosuppression;
- (viii) improved ability in rescuing tumor antigen escape;
- (ix) ability to stabilize tumor antigen; and
- (x) ability to avoid fratricide,

in comparison to its counterpart primary cell obtained from peripheral blood, umbilical cord blood, or any other donor tissues.

23. The cell or population thereof of claim 21, wherein the cell further comprises a high affinity non-cleavable CD16 (hnCD16) or a variant thereof.

24. The cell or population thereof of claim 21, wherein the CD16 or a variant thereof comprises at least one of:

- (a) F176V and S197P in ectodomain domain of CD16;

- (b) a full or partial ectodomain originated from CD64;
 - (c) a non-native (or non-CD16) transmembrane domain;
 - (d) a non-native (or non-CD16) intracellular domain;
 - (e) a non-native (or non-CD16) signaling domain;
 - (f) a non-native stimulatory domain; and
 - (g) transmembrane, signaling, and stimulatory domains that are not originated from CD16, and are originated from a same or different polypeptide.
25. The cell or population thereof of claim 24, wherein:
- (a) the non-native transmembrane domain is derived from CD3D, CD3E, CD3G, CD3 ζ , CD4, CD8, CD8a, CD8b, CD27, CD28, CD40, CD84, CD166, 4-1BB, OX40, ICOS, ICAM-1, CTLA-4, PD-1, LAG-3, 2B4, BTLA, CD16, IL7, IL12, IL15, KIR2DL4, KIR2DS1, NKp30, NKp44, NKp46, NKG2C, NKG2D, CS1, or T cell receptor (TCR) polypeptide;
 - (b) the non-native stimulatory domain is derived from CD27, CD28, 4-1BB, OX40, ICOS, PD-1, LAG-3, 2B4, BTLA, DAP10, DAP12, CTLA-4, or NKG2D polypeptide;
 - (c) the non-native signaling domain is derived from CD3 ζ , 2B4, DAP10, DAP12, DNAM1, CD137 (41BB), IL21, IL7, IL12, IL15, NKp30, NKp44, NKp46, NKG2C, or NKG2D polypeptide; or
 - (d) the non-native transmembrane domain is derived from NKG2D, the non-native stimulatory domain is derived from 2B4, and the non-native signaling domain is derived from CD3 ζ .
26. The cell or population thereof of claim 21, wherein the cell further comprises a second CAR, and wherein the second CAR is:
- (i) T cell specific or NK cell specific;
 - (ii) bi-specific antigen binding CAR;
 - (iii) a switchable CAR;
 - (iv) a dimerized CAR;
 - (v) a split CAR;
 - (vi) a multi-chain CAR;
 - (vii) an inducible CAR;
 - (viii) co-expressed with a partial or full peptide of a cell surface expressed exogenous cytokine and/or a receptor thereof, optionally in separate constructs or in a bi-cistronic construct;

- (xi) co-expressed with a checkpoint inhibitor, optionally in separate constructs or in a bi-cistronic construct;
- (xii) specific to at least one of CD19, BCMA, CD20, CD22, CD38, CD123, HER2, CD52, EGFR, GD2, MICA/B, MSLN, VEGF-R2, PSMA and PDL1; and/or
- (xiii) specific to any one of ADGRE2, carbonic anhydrase IX (CAIX), CCR1, CCR4, carcinoembryonic antigen (CEA), CD3, CD5, CD7, CD8, CD10, CD20, CD22, CD30, CD33, CD34, CD38, CD41, CD44, CD44V6, CD49f, CD56, CD70, CD74, CD99, CD123, CD133, CD138, , CDS, CLEC12A, an antigen of a cytomegalovirus (CMV) infected cell, epithelial glycoprotein2 (EGP 2), epithelial glycoprotein-40 (EGP-40), epithelial cell adhesion molecule (EpCAM), EGFRvIII, receptor tyrosine-protein kinases erb- B2,3,4, EGFR, EGFR-VIII, ERBB folate-binding protein (FBP), fetal acetylcholine receptor (AChR), folate receptor-a, Ganglioside G2 (GD2), Ganglioside G3 (GD3), human Epidermal Growth Factor Receptor 2 (HER-2), human telomerase reverse transcriptase (hTERT), ICAM-1, Integrin B7, Interleukin-13 receptor subunit alpha-2 (IL-13R α 2), κ -light chain, kinase insert domain receptor (KDR), Lewis A (CA19.9), Lewis Y (LeY), L1 cell adhesion molecule (L1-CAM), LILRB2, melanoma antigen family A 1 (MAGE-A1), MICA/B, Mucin 1 (Muc-1), Mucin 16 (Muc-16), Mesothelin (MSLN), NKCSI, NKG2D ligands, c-Met, cancer-testis antigen NY-ESO-1, oncofetal antigen (h5T4), PRAME, prostate stem cell antigen (PSCA), PRAME prostate-specific membrane antigen (PSMA), tumor-associated glycoprotein 72 (TAG-72), TIM-3, TRBC1, TRBC2, vascular endothelial growth factor R2 (VEGF- R2), Wilms tumor protein (WT-1), and a pathogen antigen.

27. The cell or population thereof of claim 21, wherein the cell comprises a partial or full peptide of a cell surface expressed exogenous cytokine and/or a receptor thereof, wherein the exogenous cytokine or receptor thereof:

- (a) comprises at least one of IL2, IL4, IL6, IL7, IL9, IL10, IL11, IL12, IL15, IL18, IL21, and its respective receptor(s); or
- (b) comprises at least one of:
 - (i) co-expression of IL15 and IL15R α by using a self-cleaving peptide;
 - (ii) a fusion protein of IL15 and IL15R α ;
 - (iii) an IL15/IL15R α fusion protein with intracellular domain of IL15R α truncated or eliminated;
 - (iv) a fusion protein of IL15 and membrane bound Sushi domain of IL15R α ;

- (v) a fusion protein of IL15 and IL15R β ;
 - (vi) a fusion protein of IL15 and common receptor γ C, wherein the common receptor γ C is native or modified; and
 - (vii) a homodimer of IL15R β ,
- wherein any one of (i)-(vii) can be co-expressed with a CAR in separate constructs or in a bi-cistronic construct;
- and optionally,
- (c) is transiently expressed.

28. The cell or population thereof of claim 19, wherein the derivative effector cell is capable of recruiting, and/or migrating T cells to tumor sites, and wherein the derivative effector cell is capable of reducing tumor immunosuppression in the presence of one or more checkpoint inhibitors.

29. The cell or population thereof of claim 26 or 28, wherein the checkpoint inhibitors are antagonists to one or more checkpoint molecules comprising: PD-1, PDL-1, TIM-3, TIGIT, LAG-3, CTLA-4, 2B4, 4-1BB, 4-1BBL, A2aR, BATE, BTLA, CD39, CD47, CD73, CD94, CD96, CD160, CD200, CD200R, CD274, CEACAM1, CSF-1R, Foxp1, GARP, HVEM, IDO, EDO, TDO, LAIR-1, MICA/B, NR4A2, MAEB, OCT-2, Rara (retinoic acid receptor alpha), TLR3, VISTA, NKG2A/HLA-E, and inhibitory KIR.

30. The cell or population thereof of claim 29, wherein the checkpoint inhibitors comprise:
- (a) one or more of atezolizumab, avelumab, durvalumab, ipilimumab, IPH4102, IPH43, IPH33, liriumab, monalizumab, nivolumab, pembrolizumab, and their derivatives or functional equivalents; or
 - (b) at least one of atezolizumab, nivolumab, and pembrolizumab.

31. The cell or population thereof of claim 20, wherein the derivative effector cell comprises a derivative CD34 cell, a derivative hematopoietic stem and progenitor cell, a derivative hematopoietic multipotent progenitor cell, a derivative T cell progenitor, a derivative NK cell progenitor, a derivative T cell, a derivative NKT cell, a derivative NK cell, a derivative B cell, or a derivative immune effector cell.

32. The cell or population thereof of claim 19, wherein the cell comprises:
- (i) one or more exogenous polynucleotides integrated in a safe harbor locus or a selected gene locus; or

- (ii) more than two exogenous polynucleotides integrated in different safe harbor loci or two or more selected gene loci.

33. The cell or population thereof of claim 32, wherein the safe harbor locus comprises at least one of AAVS1, CCR5, ROSA26, collagen, HTRP, H11, GAPDH, or RUNX1; and wherein the selected gene locus is one of B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR, NKG2A, NKG2D, CD38, CD25, CD69, CD44, CD58, CD54, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, or TIGIT; and wherein the integration of the exogenous polynucleotides knocks out expression of the gene in the locus.

34. The cell or population thereof of claim 33, wherein the TCR locus is a constant region of TCR alpha or TCR beta.

35. A composition comprising the cell or population thereof of any one of the claims 19-34.

36. A composition for therapeutic use comprising the derivative effector cell of any one of the claims 19-34, and one or more therapeutic agents.

37. The composition of claim 36, wherein the one or more therapeutic agents comprise a peptide, a cytokine, a checkpoint inhibitor, a mitogen, a growth factor, a small RNA, a dsRNA (double stranded RNA), mononuclear blood cells, feeder cells, feeder cell components or replacement factors thereof, a vector comprising one or more polynucleic acids of interest, an antibody or functional variant or fragment thereof, a chemotherapeutic agent or a radioactive moiety, or an immunomodulatory drug (IMiD).

38. The composition of claim 37, wherein

- (1) the checkpoint inhibitor comprises:
- (a) one or more antagonists to checkpoint molecules comprising PD-1, PDL-1, TIM-3, TIGIT, LAG-3, CTLA-4, 2B4, 4-1BB, 4-1BBL, A2aR, BATE, BTLA, CD39, CD47, CD73, CD94, CD96, CD160, CD200, CD200R, CD274, CEACAM1, CSF-1R, Foxp1, GARP, HVEM, IDO, EDO, TDO, LAIR-1, MICA/B, NR4A2, MAFB, OCT-2, Rara (retinoic acid receptor alpha), TLR3, VISTA, NKG2A/HLA-E, or inhibitory KIR;
 - (b) one or more of atezolizumab, avelumab, durvalumab, ipilimumab, IPH4102, IPH43, IPH33, lirimumab, monalizumab, nivolumab, pembrolizumab, and their derivatives or functional equivalents; or
 - (c) at least one of atezolizumab, nivolumab, and pembrolizumab; or

- (2) the one or more therapeutic agents comprise one or more of venetoclax, azacitidine, and pomalidomide.
39. The composition of claim 37, wherein the antibody comprises:
- (a) anti-CD20, anti-HER2, anti-CD52, anti-EGFR, anti-CD123, anti-GD2, anti-PDL1, and/or anti-CD38 antibody;
 - (b) one or more of rituximab, veltuzumab, ofatumumab, ublituximab, ocaratuzumab, obinutuzumab, trastuzumab, pertuzumab, alemtuzumab, certuximab, dinutuximab, avelumab, daratumumab, isatuximab, MOR202, 7G3, CSL362, elotuzumab, and their humanized or Fc modified variants or fragments and their functional equivalents and biosimilars; or
 - (c) daratumumab, and wherein the derivative effector cells comprise a CD38 knockout, and optionally an expression of CD16 or a variant thereof.
40. Therapeutic use of the composition of any one of the claims 35-39 by introducing the composition to a subject suitable for adoptive cell therapy, wherein the subject has an autoimmune disorder, a hematological malignancy, a solid tumor, cancer, or a viral infection.
41. A method of manufacturing a derivative effector cell comprising the CAR of claim 1, wherein the method comprises differentiating a genetically engineered iPSC, wherein the iPSC comprises a polynucleotide encoding the CAR, and optionally one or more editing resulting in:
- (i) CD38 knockout;
 - (ii) B2M null or low, and optionally CIITA null or low, in comparison to its counterpart primary cell;
 - (iii) introduced expression of HLA-G or non-cleavable HLA-G, or knockout in one or both of CD58 and CD54;
 - (iv) a CD16 or a variant thereof;
 - (v) a chimeric antigen receptor (CAR) with a different targeting specificity;
 - (vi) a partial or full peptide of a cell surface expressed exogenous cytokine or a receptor thereof;
 - (vii) at least one of the genotypes listed in Table 2;
 - (viii) deletion or reduced expression in at least one of TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR α or β constant region, NKG2A, NKG2D, CD25, CD69, CD44, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, and TIGIT, in comparison to its counterpart primary cell; and/or

- (ix) introduced or increased expression in at least one of HLA-E, 41BBL, CD3, CD4, CD8, CD16, CD47, CD113, CD131, CD137, CD80, PDL1, A_{2A}R, antigen-specific TCR, Fc receptor, an engager, and surface triggering receptor for coupling with bi- or multi- specific or universal engagers, in comparison to its counterpart primary cell.
42. The method of claim 41, further comprising genomically engineering a clonal iPSC to knock in a polynucleotide encoding the CAR; and optionally:
- (i) to knock out CD38,
 - (ii) to knock out B2M and CIITA,
 - (iii) to knock out one or both CD58 and CD54, and/or
 - (iv) to introduce expression of HLA-G or non-cleavable HLA-G, a high affinity non-cleavable CD16 or a variant thereof, a second CAR, and/or a partial or full peptide of a cell surface expressed exogenous cytokine or a receptor thereof.
43. The method of claim 42, wherein the genomic engineering comprises targeted editing.
44. The method of claim 43, wherein the targeted editing comprises deletion, insertion, or in/del, and wherein the targeted editing is carried out by CRISPR, ZFN, TALEN, homing nuclease, homology recombination, or any other functional variation of these methods.
45. CRISPR mediated editing of clonal iPSCs, wherein the editing comprises a knock-in of a polynucleotide encoding the CAR of claim 1.
46. The CRISPR mediated editing of claim 45:
- (a) wherein the editing of clonal iPSCs further comprises knocking out CD38, or
 - (b) wherein the CAR is inserted at one of the gene loci comprising: B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR α or β constant region, NKG2A, NKG2D, CD38, CD25, CD69, CD44, CD58, CD54, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, or TIGIT; and wherein the insertion knocks out expression of the gene in the locus.
47. A method of treating a disease or a condition comprising administering to a subject in need thereof cells comprising the CAR of any one of claims 1-18.
48. The method of claim 47, wherein the cells comprise derivative effector cells comprising a CD38 knockout, a CD16 or a variant thereof, and optionally comprise:

- (i) B2M and CIITA knockout;
 - (ii) introduced expression of HLA-G or non-cleavable HLA-G, or knockout of one or both of CD58 and CD54;
 - (iii) introduced expression of a second CAR, and/or a partial or full peptide of a cell surface expressed exogenous cytokine or a receptor thereof; and/or
 - (iii) at least one of the genotypes listed in Table 2.
49. The method of claim 47, wherein administration of the cells results in one or more of:
- (i) reducing tumor cell surface shedding of MICA/B antigen;
 - (ii) increasing tumor cell surface MICA/B density;
 - (iii) preventing tumor antigen escape;
 - (iv) overcoming tumor microenvironment suppression;
 - (v) enhancing effector cell activation and killing function; and
 - (vi) *in vivo* tumor progression control, tumor cell burden reduction, tumor clearance, and/or improving rate of survival;
- as compared to treatment using effector cells without the CAR of claim 1.

CLAIMS

1. A chimeric antigen receptor comprising: an ectodomain comprising at least one antigen recognition domain; a transmembrane domain; and an endodomain comprising at least one signaling domain;

5 wherein the at least one signaling domain is originated from a cytoplasmic domain of a signal transducing protein specific to T and/or NK cell activation or functioning;

in which the chimeric antigen receptor, when comprised in an induced pluripotent stem cell (iPSC), promotes differentiation of the iPSC directed to a desired derivative effector cell, and in which the iPSC-derived effector cell differentiated from the iPSC has at least
10 one of the following characteristics comprising:

(i) improved persistency and/or survival;

(ii) improved cell expansion;

(iii) increased cytotoxicity;

(iv) increased resistance to allorejection;

15 (v) improved tumor penetration;

(vi) enhanced ability in migrating, and/or activating or recruiting bystander immune cells, to tumor sites; and

(vii) enhanced ability to reduce tumor immunosuppression,

in comparison to a primary immune cell obtained from peripheral blood, umbilical cord
20 blood, or any other donor tissues.

2. A chimeric antigen receptor according to Claim 1, in which

(a) the signal transducing protein comprises any one of: 2B4 (Natural killer Cell Receptor 2B4), 4-1BB (Tumor necrosis factor receptor superfamily member 9), CD16 (IgG Fc region Receptor III-A), CD2 (T-cell surface antigen CD2), CD28 (T-cell-specific surface glycoprotein CD28), CD28H (Transmembrane and immunoglobulin domain-containing protein 2), CD3 ζ (T-cell surface glycoprotein CD3 zeta chain), DAP10 (Hematopoietic cell signal transducer), DAP12 (TYRO protein tyrosine kinase-binding protein), DNAM1 (CD226 antigen), Fc ϵ RI γ (High affinity immunoglobulin epsilon receptor subunit gamma), IL21R (Interleukin-21 receptor), IL-2R β /IL-15RB
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(Interleukin-2 receptor subunit beta), IL-2R γ (Cytokine receptor common subunit gamma), IL-7R (Interleukin-7 receptor subunit alpha), KIR2DS2 (Killer cell immunoglobulin-like receptor 2DS2), NKG2D (NKG2-D type II integral membrane protein), NKp30 (Natural cytotoxicity triggering receptor 3), NKp44 (Natural cytotoxicity triggering receptor 2), NKp46 (Natural cytotoxicity triggering receptor 1), CS1 (SLAM family member 7), and CD8 (T-cell surface glycoprotein CD8 alpha chain); and/or

(b) the at least one signaling domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of 2B4, 4-1BB, CD16, CD2, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ IL21R, IL-2R β (IL-15R β), IL-2R γ , IL-7R, KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CD3 ζ 1XX, CS1, or CD8, represented by SEQ ID NOs: 21-41, 54 and 56, respectively.

3. A chimeric antigen receptor according to Claim 2, in which the at least one signaling domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of 2B4, 4-1BB, CD16, CD2, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ IL21R, IL-2R β (IL-15R β), IL-2R γ , IL-7R, KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CD3 ζ 1XX, CS1, or CD8, represented by SEQ ID NOs: 21-41, 54 and 56, respectively; and in which the portion of the cytoplasmic domain comprises an ITAM (immunoreceptor tyrosine-based activation motif), a YxxM motif, a TxYxxV/I motif, FcR γ , a hemi-ITAM, and/or an ITT-like motif.

4. A chimeric antigen receptor according to Claim 1, in which the endodomain comprises a first signaling domain, a second signaling domain, and optionally a third signaling domain; and in which the first, second and third signaling domains are different.

5. A chimeric antigen receptor according to Claim 4, in which the second or the third signaling domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of 2B4, 4-1BB, CD16, CD2, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ IL21R, IL-2R β (IL-15R β), IL-2R γ , IL-7R, KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CD3 ζ 1XX, CS1, or CD8, represented by SEQ ID NOs: 21-41, 54 and 56, respectively.

6. A chimeric antigen receptor according to Claim 1, in which the endodomain comprises only one signaling domain, in which the endodomain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to the cytoplasmic domain, or a portion thereof, of DNAM1, CD28H, KIR2DS2, DAP12 or DAP10.
7. A chimeric antigen receptor according to Claim 4, in which the endodomain comprises two different signaling domains, and in which the endodomain domain comprises fused cytoplasmic domains, or portions thereof, in any one of the forms: 2B4-CD3 ζ /1XX, 2B4-DNAM1, 2B4-FcERI γ , 2B4-DAP10, CD16-DNAM1, CD16-DAP10, CD16-DAP12, CD2-CD3 ζ /1XX, CD2-DNAM1, CD2-FcERI γ , CD2-DAP10, CD28-DNAM1, CD28-FcERI γ , CD28-DAP10, CD28-DAP12, CD28H-CD3 ζ /1XX, DAP10-CD3 ζ /1XX, DAP10-DAP12, DAP12-CD3 ζ /1XX, DAP12-DAP10, DNAM1-CD3 ζ /1XX, KIR2DS2-CD3 ζ /1XX, KIR2DS2-DAP10, KIR2DS2-2B4, and NKp46-2B4.
8. A chimeric antigen receptor according to Claim 4, in which the endodomain comprises three different signaling domains, and in which the endodomain domain comprises fused cytoplasmic domains, or portions thereof, in any one of the forms comprising: 2B4-DAP10-CD3 ζ /1XX, 2B4-IL21R-DAP10, 2B4-IL2RB-DAP10, 2B4-IL2RB-CD3 ζ /1XX, 2B4-41BB-DAP10, CD16-2B4-DAP10, and KIR2DS2-2B4-CD3 ζ /1XX.
9. A chimeric antigen receptor according to Claim 1, in which the transmembrane domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to a transmembrane region, or a portion thereof, of CD2, CD3D, CD3E, CD3G, CD3 ζ , CD4, CD8, CD8a, CD8b, CD16, CD27, CD28, CD28H, CD40, CD84, CD166, 4-1BB, OX40, ICOS, ICAM-1, CTLA4, PD1, LAG3, 2B4, BTLA, DNAM1, DAP10, DAP12, FcERI γ , IL7, IL12, IL15, KIR2DL4, KIR2DS1, KIR2DS2, NKp30, NKp44, NKp46, NKG2C, NKG2D, CS1, or T cell receptor polypeptide.
10. A chimeric antigen receptor according to Claim 1, in which the transmembrane domain comprises an amino acid sequence that has at least about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, or about 99% identity to a transmembrane region, or a portion thereof, of 2B4, CD2, CD16, CD28, CD28H, CD3 ζ , DAP10, DAP12, DNAM1, FcERI γ , KIR2DS2, NKG2D, NKp30, NKp44, NKp46, CS1, or CD8, represented by SEQ ID NOs: 1-20, 53 and 55, respectively.

11. A chimeric antigen receptor according to Claim 1, in which the transmembrane domain and its immediately linked signaling domain are from a same protein or from different proteins.

12. A chimeric antigen receptor according to Claim 1, in which the chimeric antigen receptor comprises a transmembrane domain and an endodomain (*TM*-(endodomain)), and in which the chimeric antigen receptor comprises:

(i) one of the forms: *NKG2D*-(2B4-IL2RB-CD3 ζ), *CD8*-(41BB- CD3 ζ 1XX), *CD28*-(CD28-2B4-CD3 ζ), *CD28H*-(CD28H-CD3 ζ), *DNAM1*-(DNAM1-CD3 ζ), *DAP10*-(DAP10-CD3 ζ), *KIR2DS2*-(KIR2DS2-CD3 ζ), *KIR2DS2*-(KIR2DS2-DAP10),
KIR2DS2-(KIR2DS2-2B4), *CD16*-(CD16-2B4-DAP10), *CD16*-(CD16-DNAM1),
NKp46-(NKp46-2B4), *NKp46*-(NKp46-2B4-CD3 ζ), *NKp46*-(NKp46-CD2-Dap10),
CD2-(CD2-CD3 ζ), *2B4*-(2B4-CD3 ζ), *2B4*-(2B4-FcERIg), and *CS1*-(CS1-CD3 ζ); or

(ii) an amino acid sequence having about 85%, about 90%, about 95%, about 96%, about 97%, about 98%, about 99%, or 100% identity to a sequence represent by each of
 SEQ ID NOs: 57-74.

13. A chimeric antigen receptor according to Claim 1, in which the antigen recognition domain specifically binds an antigen associated with a disease, a pathogen, a liquid tumor, or a solid tumor.

14. A chimeric antigen receptor according to Claim 1, in which the antigen recognition domain is specific to:

(i) any one of CD19, BCMA, CD20, CD22, CD38, CD123, HER2, CD52, EGFR, GD2, MICA/B, MSLN, VEGF-R2, PSMA and PDL1; or

(ii) any one of ADGRE2, carbonic anhydrase IX (CAIX), CCRI, CCR4, carcinoembryonic antigen (CEA), CD3, CD5, CD7, CD8, CD10, CD20, CD22, CD30, CD33, CD34, CD38, CD41, CD44, CD44V6, CD49f, CD56, CD70, CD74, CD99, CD123, CD133, CD138, CDS, CLEC12A, an antigen of a cytomegalovirus (CMV) infected cell, epithelial glycoprotein2 (EGP-2), epithelial glycoprotein-40 (EGP-40), epithelial cell adhesion molecule (EpCAM), EGFRvIII, receptor tyrosine-protein kinases erb- B2,3,4, EGFR, EGFR-VIII, ERBB folate-binding protein (FBP), fetal acetylcholine receptor (AChR), folate receptor-a, Ganglioside G2 (GD2), Ganglioside

G3 (GD3), human Epidermal Growth Factor Receptor 2 (HER2), human telomerase reverse transcriptase (hTERT), ICAM-1, Integrin B7, Interleukin-13 receptor subunit alpha-2 (IL-13R α 2), κ -light chain, kinase insert domain receptor (KDR), Lewis A (CA19.9), Lewis Y (LeY), L1 cell adhesion molecule (L1-CAM), LILRB2, melanoma antigen family A 1 (MAGE-A1), MICA/B, Mucin 1 (Muc-1), Mucin 16 (Muc-16), Mesothelin (MSLN), NKCSI, NKG2D ligands, c-Met, cancer-testis antigen NY-ESO-1, oncofetal antigen (h5T4), PRAME, prostate stem cell antigen (PSCA), PRAME prostate-specific membrane antigen (PSMA), tumor- associated glycoprotein 72 (TAG-72), TIM-3, TRBC1, TRBC2, vascular endothelial growth factor R2 (VEGF-R2), Wilms tumor protein (WT-1), and a pathogen antigen.

15. A chimeric antigen receptor according to Claim 1, in which the ectodomain comprises one or more of:

- (i) two antigen recognition domains;
- (ii) a signal peptide; and/or
- (iii) a spacer/hinge.

16. A chimeric antigen receptor according to Claim 1, in which the chimeric antigen receptor is comprised in a bi-cistronic construct co-expressing a partial or full length peptide of a cell surface expressed exogenous cytokine or a receptor thereof, in which the exogenous cytokine or receptor thereof comprises:

- (a) at least one of IL2, IL4, IL6, IL7, IL9, IL10, IL11, IL12, IL15, IL18, IL21, and its respective receptor(s); or
- (b) at least one of:
 - (i) co-expression of IL15 and IL15R α by using a self-cleaving peptide;
 - (ii) a fusion protein of IL15 and IL15R α ;
 - (iii) an IL15/IL15R α fusion protein with intracellular domain of IL15R α truncated or eliminated;
 - (iv) a fusion protein of IL15 and membrane bound Sushi domain of IL15R α ;
 - (v) a fusion protein of IL15 and IL15R β ;

(vi) a fusion protein of IL15 and common receptor γ C, in which the common receptor γ C is native or modified; and

(vii) a homodimer of IL15R β .

17. A chimeric antigen receptor according to Claim 1, in which the derivative effector cell from iPSC differentiation comprises one or more of: a derivative CD34 cell, a derivative hematopoietic stem and progenitor cell, a derivative hematopoietic multipotent progenitor cell, a derivative T cell progenitor, a derivative NK cell progenitor, a derivative T cell, a derivative NKT cell, a derivative NK cell, a derivative B cell, or a derivative immune effector cell.

18. A chimeric antigen receptor according to Claim 17, in which the iPSC-derived immune effector cell expresses the chimeric antigen receptor, and comprises at least one functional feature that is not present in a primary T, NK, and/or NKT cell.

19. A cell or a population thereof, in which:

(i) the cell is an immune cell, an induced pluripotent cell (iPSC), a clonal iPSC, or an iPS cell line cell; or the cell is a derivative effector cell obtained from differentiating the iPSC; and

(ii) the cell comprises at least one chimeric antigen receptor (CAR) according to any preceding claim.

20. A cell or population thereof according to Claim 19, in which the derivative effector cell is a hematopoietic cell, and comprises longer telomeres in comparison to its counterpart primary cell obtained from peripheral blood, umbilical cord blood, or any other donor tissues; or in which the CAR has at least one of the following characteristics:

(i) being T or NK cell specific;

(ii) bi-specific in antigen binding;

(iii) being a switchable CAR;

(iv) being a dimerized CAR;

(v) being a split CAR;

(vi) being a multi-chain CAR;

(vii) being an inducible CAR; and

(viii) being inserted at one of the following gene loci: B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR α or β constant region, NKG2A, NKG2D, CD38, CD25, CD69, CD44, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, or TIGIT, in which the insertion knocks out or reduces expression of the gene in the locus.

21. A cell or population thereof according to Claim 19, in which the cell further comprises one or more of:

(i) CD38 knockout;

(ii) HLA-I deficiency, and optionally HLA-II deficiency, in comparison to its counterpart primary cell;

(iii) introduction of HLA-G or non-cleavable HLA-G;

(iv) a CD16 or a variant thereof;

(v) a second CAR having different targeting specificity;

(vi) a partial or full peptide of a cell surface expressed exogenous cytokine and/or a receptor thereof;

(vii) at least one of the genotypes listed in Table 2;

(viii) deletion or disruption in at least one of B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR, NKG2A, NKG2D, CD25, CD69, CD44, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, and TIGIT, in comparison to its counterpart primary cell; or

(ix) introduction or upregulation of at least one of HLA-E, 41BBL, CD3, CD4, CD8, CD16, CD47, CD113, CD131, CD137, CD80, PDL1, A_{2A}R, antigen-specific TCR, Fc receptor, an engager, and surface triggering receptor for coupling with an agonist, in comparison to its counterpart primary cell.

22. A cell or population thereof according to Claim 19, in which the cell is a derivative effector cell, and has at least one of the following characteristics comprising:

(i) improved persistency and/or survival;

(ii) increased resistance to alloreactive recipient immune cells;

- (iii) increased cytotoxicity;
- (iv) improved tumor penetration;
- (v) enhanced or acquired ADCC;
- (vi) enhanced ability in migrating, and/or activating or recruiting bystander immune cells, to tumor sites;
- (vii) enhanced ability to reduce tumor immunosuppression;
- (viii) improved ability in rescuing tumor antigen escape;
- (ix) ability to stabilize tumor antigen; and
- (x) ability to avoid fratricide,

10 in comparison to its counterpart primary cell obtained from peripheral blood, umbilical cord blood, or any other donor tissues.

23. A cell or population thereof according to Claim 21, in which the cell further comprises a high affinity non-cleavable CD16 (hnCD16) or a variant thereof.

15 24. A cell or population thereof according to Claim 21, in which the CD16 or a variant thereof comprises at least one of:

- (a) F176V and S197P in ectodomain domain of CD16;
- (b) a full or partial ectodomain originated from CD64;
- (c) a non-native (or non-CD16) transmembrane domain;
- (d) a non-native (or non-CD16) intracellular domain;
- 20 (e) a non-native (or non-CD16) signaling domain;
- (f) a non-native stimulatory domain; and
- (g) transmembrane, signaling, and stimulatory domains that are not originated from CD16, and are originated from a same or different polypeptide.

25. A cell or population thereof according to Claim 24, in which:

25 (a) the non-native transmembrane domain is derived from CD3D, CD3E, CD3G, CD3ζ, CD4, CD8, CD8a, CD8b, CD27, CD28, CD40, CD84, CD166, 4-1BB, OX40, ICOS, ICAM-1, CTLA-4, PD-1, LAG-3, 2B4, BTLA, CD16, IL7, IL12, IL15,

KIR2DL4, KIR2DS1, NKp30, NKp44, NKp46, NKG2C, NKG2D, CS1, or T cell receptor (TCR) polypeptide;

(b) the non-native stimulatory domain is derived from CD27, CD28, 4-1BB, OX40, ICOS, PD-1, LAG-3, 2B4, BTLA, DAP10, DAP12, CTLA-4, or NKG2D polypeptide;

5 (c) the non-native signaling domain is derived from CD3 ζ , 2B4, DAP10, DAP12, DNAM1, CD137 (41BB), IL21, IL7, IL12, IL15, NKp30, NKp44, NKp46, NKG2C, or NKG2D polypeptide; or

(d) the non-native transmembrane domain is derived from NKG2D, the non-native stimulatory domain is derived from 2B4, and the non-native signaling domain is derived
10 from CD3 ζ .

26. A cell or population thereof according to Claim 21, in which the cell further comprises a second CAR, and in which the second CAR is:

(i) T cell specific or NK cell specific;

(ii) bi-specific antigen binding CAR;

15 (iii) a switchable CAR;

(iv) a dimerized CAR;

(v) a split CAR;

(vi) a multi-chain CAR;

(vii) an inducible CAR;

20 (viii) co-expressed with a partial or full peptide of a cell surface expressed exogenous cytokine and/or a receptor thereof, optionally in separate constructs or in a bi-cistronic construct;

(xi) co-expressed with a checkpoint inhibitor, optionally in separate constructs or in a bi-cistronic construct;

25 (xii) specific to at least one of CD19, BCMA, CD20, CD22, CD38, CD123, HER2, CD52, EGFR, GD2, MICA/B, MSLN, VEGF-R2, PSMA and PDL1; and/or

(xiii) specific to any one of ADGRE2, carbonic anhydrase IX (CAIX), CCR1, CCR4, carcinoembryonic antigen (CEA), CD3, CD5, CD7, CD8, CD10, CD20, CD22, CD30, CD33, CD34, CD38, CD41, CD44, CD44V6, CD49f, CD56, CD70, CD74, CD99,

CD123, CD133, CD138, , CDS, CLEC12A, an antigen of a cytomegalovirus (CMV) infected cell, epithelial glycoprotein2 (EGP-2), epithelial glycoprotein-40 (EGP-40), epithelial cell adhesion molecule (EpCAM), EGFRvIII, receptor tyrosine-protein kinases erb- B2,3,4, EGFR, EGFR-VIII, ERBB folate-binding protein (FBP), fetal acetylcholine receptor (AChR), folate receptor-a, Ganglioside G2 (GD2), Ganglioside G3 (GD3), human Epidermal Growth Factor Receptor 2 (HER2), human telomerase reverse transcriptase (hTERT), ICAM-1, Integrin B7, Interleukin-13 receptor subunit alpha-2 (IL-13R α 2), κ -light chain, kinase insert domain receptor (KDR), Lewis A (CA19.9), Lewis Y (LeY), L1 cell adhesion molecule (L1-CAM), LILRB2, melanoma antigen family A 1 (MAGE-A1), MICA/B, Mucin 1 (Muc-1), Mucin 16 (Muc-16), Mesothelin (MSLN), NKCSI, NKG2D ligands, c-Met, cancer-testis antigen NY-ESO-1, oncofetal antigen (h5T4), PRAME, prostate stem cell antigen (PSCA), PRAME prostate-specific membrane antigen (PSMA), tumor- associated glycoprotein 72 (TAG-72), TIM-3, TRBC1, TRBC2, vascular endothelial growth factor R2 (VEGF-R2), Wilms tumor protein (WT-1), and a pathogen antigen.

27. A cell or population thereof according to Claim 21, in which the cell comprises a partial or full peptide of a cell surface expressed exogenous cytokine and/or a receptor thereof, in which the exogenous cytokine or receptor thereof:

(a) comprises at least one of IL2, IL4, IL6, IL7, IL9, IL10, IL11, IL12, IL15, IL18, IL21, and its respective receptor(s); or

(b) comprises at least one of:

(i) co-expression of IL15 and IL15R α by using a self-cleaving peptide;

(ii) a fusion protein of IL15 and IL15R α ;

(iii) an IL15/IL15R α fusion protein with intracellular domain of IL15R α truncated or eliminated;

(iv) a fusion protein of IL15 and membrane bound Sushi domain of IL15R α ;

(v) a fusion protein of IL15 and IL15R β ;

(vi) a fusion protein of IL15 and common receptor γ C, in which the common receptor γ C is native or modified; and

(vii) a homodimer of IL15R β ,

in which any one of (i)-(vii) can be co-expressed with a CAR in separate constructs or in a bi-cistronic construct;

and optionally,

(c) is transiently expressed.

5 28. A cell or population thereof according to Claim 19, in which the derivative effector cell is capable of recruiting, and/or migrating T cells to tumor sites, and in which the derivative effector cell is capable of reducing tumor immunosuppression in the presence of one or more checkpoint inhibitors.

10 29. A cell or population thereof according to Claim 26 or 28, in which the checkpoint inhibitors are antagonists to one or more checkpoint molecules comprising: PD-1, PDL-1, TIM-3, TIGIT, LAG-3, CTLA-4, 2B4, 4-1BB, 4-1BBL, A_{2A}R, BATE, BTLA, CD39, CD47, CD73, CD94, CD96, CD160, CD200, CD200R, CD274, CEACAM1, CSF-1R, Foxp1, GARP, HVEM, IDO, EDO, TDO, LAIR-1, MICA/B, NR4A2, MAFB, OCT-2, Rara (retinoic acid receptor alpha), TLR3, VISTA, NKG2A/HLA-E, and inhibitory KIR.

15 30. A cell or population thereof according to Claim 29, in which the checkpoint inhibitors comprise:

(a) one or more of atezolizumab, avelumab, durvalumab, ipilimumab, IPH4102, IPH43, IPH33, lirimumab, monalizumab, nivolumab, pembrolizumab, and their derivatives or functional equivalents; or

20 (b) at least one of atezolizumab, nivolumab, and pembrolizumab.

31. A cell or population thereof according to Claim 20, in which the derivative effector cell comprises a derivative CD34 cell, a derivative hematopoietic stem and progenitor cell, a derivative hematopoietic multipotent progenitor cell, a derivative T cell progenitor, a derivative NK cell progenitor, a derivative T cell, a derivative NKT cell, a derivative NK cell,
25 a derivative B cell, or a derivative immune effector cell.

32. A cell or population thereof according to Claim 19, in which the cell comprises:

(i) one or more exogenous polynucleotides integrated in a safe harbor locus or a selected gene locus; or

(ii) more than two exogenous polynucleotides integrated in different safe harbor loci or two or more selected gene loci.

33. A cell or population thereof according to Claim 32, in which the safe harbor locus comprises at least one of AAVS1, CCR5, ROSA26, collagen, HTRP, H11, GAPDH, or
5 RUNX1; and in which the selected gene locus is one of B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR, NKG2A, NKG2D, CD38, CD25, CD69, CD44, , CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, or TIGIT; and in which the integration of the exogenous polynucleotides knocks out expression of the gene in the locus.

34. A cell or population thereof according to Claim 33, in which the TCR locus is a constant
10 region of TCR alpha or TCR beta.

35. A composition comprising the cell or population thereof according to any of Claims 19 to 34.

36. A composition for therapeutic use comprising the derivative effector cell according to any of Claims 19 to 34, and one or more therapeutic agents.

15 37. A composition according to Claim 36, in which the one or more therapeutic agents comprise a peptide, a cytokine, a checkpoint inhibitor, a mitogen, a growth factor, a small RNA, a dsRNA (double stranded RNA), mononuclear blood cells, feeder cells, feeder cell components or replacement factors thereof, a vector comprising one or more polynucleic acids of interest, an antibody or functional variant or fragment thereof, a chemotherapeutic agent or
20 a radioactive moiety, or an immunomodulatory drug (IMiD).

38. A composition according to Claim 37, in which

(1) the checkpoint inhibitor comprises:

(a) one or more antagonists to checkpoint molecules comprising PD-1, PDL-1, TIM-3, TIGIT, LAG-3, CTLA-4, 2B4, 4-1BB, 4-1BBL, A_{2A}R, BATE, BTLA,
25 CD39, CD47, CD73, CD94, CD96, CD160, CD200, CD200R, CD274, CEACAM1, CSF-1R, Foxp1, GARP, HVEM, IDO, EDO, TDO, LAIR-1, MICA/B, NR4A2, MAFB, OCT-2, Rara (retinoic acid receptor alpha), TLR3, VISTA, NKG2A/HLA-E, or inhibitory KIR;

(b) one or more of atezolizumab, avelumab, durvalumab, ipilimumab, IPH4102, IPH43, IPH33, lirimumab, monalizumab, nivolumab, pembrolizumab, and their derivatives or functional equivalents; or

(c) at least one of atezolizumab, nivolumab, and pembrolizumab; or

5 (2) the one or more therapeutic agents comprise one or more of venetoclax, azacitidine, and pomalidomide.

39. A composition according to Claim 37, in which the antibody comprises:

(a) anti-CD20, anti-HER2, anti-CD52, anti-EGFR, anti-CD123, anti-GD2, anti-PDL1, and/or anti-CD38 antibody;

10 (b) one or more of rituximab, veltuzumab, ofatumumab, ublituximab, ocaratuzumab, obinutuzumab, trastuzumab, pertuzumab, alemtuzumab, certuximab, dinutuximab, avelumab, daratumumab, isatuximab, MOR202, 7G3, CSL362, elotuzumab, and their humanized or Fc modified variants or fragments and their functional equivalents and biosimilars; or

15 (c) daratumumab, and in which the derivative effector cells comprise a CD38 knockout, and optionally an expression of CD16 or a variant thereof.

40. Use of a composition according to any of Claims 35 to 39 in the manufacture of a medicament for the treatment of an autoimmune disorder, a hematological malignancy, a solid tumor, cancer, or a viral infection.

20 41. A method of manufacturing a derivative effector cell comprising the CAR according to Claim 1, in which the method comprises differentiating a genetically engineered iPSC, in which the iPSC comprises a polynucleotide encoding the CAR, and optionally one or more editing resulting in:

(i) CD38 knockout;

25 (ii) HLA-I deficiency, and optionally HLA-II deficiency, in comparison to its counterpart primary cell;

(iii) introduction of HLA-G or non-cleavable HLA-G;

(iv) a CD16 or a variant thereof;

(v) a chimeric antigen receptor (CAR) with a different targeting specificity;

(vi) a partial or full peptide of a cell surface expressed exogenous cytokine or a receptor thereof;

(vii) at least one of the genotypes listed in Table 2;

5 (viii) deletion or disruption in at least one of B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR α or β constant region, NKG2A, NKG2D, CD25, CD69, CD44, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, and TIGIT, in comparison to its counterpart primary cell; and/or

10 (ix) introduction or upregulation of at least one of HLA-E, 41BBL, CD3, CD4, CD8, CD16, CD47, CD113, CD131, CD137, CD80, PDL1, A_{2A}R, antigen-specific TCR, Fc receptor, an engager, and surface triggering receptor for coupling with bi- or multi-specific or universal engagers, in comparison to its counterpart primary cell.

42. A method according to Claim 41, further comprising genomically engineering a clonal iPSC to knock in a polynucleotide encoding the CAR; and optionally:

(i) to knock out CD38,

15 (ii) to knock out B2M or CIITA, and/or

(iii) to introduce polynucleotides encoding HLA-G or non-cleavable HLA-G, a high affinity non-cleavable CD16 or a variant thereof, a second CAR, and/or a partial or full peptide of a cell surface expressed exogenous cytokine or a receptor thereof.

20 43. A method according to Claim 42, in which the genomic engineering comprises targeted editing.

44. A method according to Claim 43, in which the targeted editing comprises deletion, insertion, or in/del, and in which the targeted editing is carried out by CRISPR, ZFN, TALEN, homing nuclease, homology recombination, or any other functional variation of these methods.

25 45. CRISPR mediated editing of clonal iPSCs, in which the editing comprises a knock-in of a polynucleotide encoding a CAR according to Claim 1.

46. CRISPR mediated editing according to Claim 45:

(a) in which the editing of clonal iPSCs further comprises knocking out CD38, or

(b) in which the CAR is inserted at one of the gene loci comprising: B2M, TAP1, TAP2, Tapasin, NLRC5, CIITA, RFXANK, RFX5, RFXAP, TCR α or β constant region, NKG2A, NKG2D, CD38, CD25, CD69, CD44, CD56, CIS, CBL-B, SOCS2, PD1, CTLA4, LAG3, TIM3, or TIGIT; and in which the insertion knocks out expression of the gene in the locus.

47. Use of cells comprising a CAR according to any of Claims 1 to 18 in the manufacture of a medicament.

48. Use according to Claim 47, in which the cells comprise derivative effector cells comprising a CD38 knockout, a CD16 or a variant thereof, and optionally comprise:

- (i) B2M or CIITA knockout;
- (ii) introduced expression of HLA-G or non-cleavable HLA-G;
- (iii) introduction of a second CAR, and/or a partial or full peptide of a cell surface expressed exogenous cytokine or a receptor thereof; and/or
- (iii) at least one of the genotypes listed in Table 2.

49. Use according to Claim 47, in which administration of the cells results in one or more of:

- (i) reducing tumor cell surface shedding of MICA/B antigen;
- (ii) increasing tumor cell surface MICA/B density;
- (iii) preventing tumor antigen escape;
- (iv) overcoming tumor microenvironment suppression;
- (v) enhancing effector cell activation and killing function; and
- (vi) *in vivo* tumor progression control, tumor cell burden reduction, tumor clearance, and/or improving rate of survival;

as compared to treatment using effector cells without the CAR according to Claim 1.