CLAIMS

1. An isolated MAGEA1 (EADPTGHSY) peptide-MHC (HLA-A*01) (pMHC) complex binding protein comprising:

a) a T cell receptor (TCR) alpha chain variable domain comprising CDR1 having residues 46-51 of SEQ ID NO: 20, CDR2 having residues 69-75 of SEQ ID NO: 20, and CDR3 having residues 109-119 of SEQ ID NO: 20; and

b) a TCR beta chain variable domain comprising CDR1 having residues 46-50 of SEQ ID NO: 22, CDR2 having residues 68-73 of SEQ ID NO: 22, and CDR3 having residues 110-122 of SEQ ID NO: 22.

2. The binding protein of claim 1, wherein the binding protein further comprises:

1) a) a TCR alpha chain variable ($V_\alpha$) domain comprising a sequence with at least 80%, at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% identity to residues 1-130 of SEQ ID NO: 20; and

b) a TCR beta chain variable ($V_\beta$) domain comprising a sequence with at least 80%, at least 85%, at least 90, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% identity to residues 1-132 of SEQ ID NO: 22;

2) a) a TCR alpha chain comprising a sequence with at least 80%, at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% identity to the TCR alpha chain sequence of SEQ ID NO: 20; and

b) a TCR beta chain comprising a sequence with at least 80%, at least 85%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98%, at least 99%, or 100% identity to the TCR beta chain sequence of SEQ ID NO: 22;

3) a) a TCR alpha chain variable ($V_\alpha$) domain comprising residues 1-130 of SEQ ID NO: 20; and

b) a TCR beta chain variable ($V_\beta$) domain comprising residues 1-132 of SEQ ID NO: 22; or

4) a) a TCR alpha chain comprising the sequence of SEQ ID NO: 20; and

b) a TCR beta chain comprising the sequence of SEQ ID NO: 22;

3. The binding protein of claim 1 or 2, wherein:

1) the TCR $V_\alpha$ domain or TCR alpha chain is encoded by TRAV21 and/or TRAJ47 genes;

2) the TCR $V_\beta$ domain or TCR beta chain is encoded by TRBV5-4 and/or TRBJ2-7 genes;

3) the binding protein is chimeric, humanized, or human;

4) the binding protein is an antibody, an antigen-binding fragment of an antibody, a TCR, an antigen-binding fragment of a TCR, a single chain TCR (scTCR), a chimeric antigen receptor (CAR), or a fusion protein comprising a TCR and an effector domain;

5) the binding protein comprises a binding domain having a transmembrane domain, and an effector domain that is intracellular;

6) the TCR alpha chain and the TCR beta chain are covalently linked, optionally wherein the TCR alpha chain and the TCR beta chain are covalently linked through a linker peptide;

7) the TCR alpha chain and/or the TCR beta chain are covalently linked to a moiety, optionally wherein the covalently linked moiety:

a) comprises an affinity tag or a label, optionally wherein the affinity tag is selected from the group consisting of CD34 enrichment tag, Glutathione-S-Transferase (GST), calmodulin binding protein (CBP), protein C tag, Myc tag, HaloTag, HA tag, Flag tag, His tag, biotin tag, and V5 tag, and/or wherein the label is a fluorescent protein; and/or

b) is selected from the group consisting of an inflammatory agent, cytokine, toxin, cytotoxic molecule, radioactive isotope, or antibody or antigen-binding fragment thereof;

8) the binding protein binds to the pMHC complex on a cell surface;

9) the MHC is an MHC multimer, optionally wherein the MHC multimer is a tetramer;

10) the MHC alpha chain is encoded by an HLA-A*01:01 allele;

11) binding of the binding protein to the MAGEA1 (EADPTGHSY) peptide-MHC (HLA-A*01) (pMHC) complex elicits an immune response, optionally wherein the immune

response is i) a T cell response and/or a CD8+ T cell response and/or ii) selected from the group consisting of T cell expansion, cytokine release, and/or cytotoxic killing;

12) the binding protein is capable of specifically and/or selectively binding to the MAGEA1 immunogenic peptide-MHC (pMHC) complex with a $K_d$ less than or equal to $1x10^{-4}$ M, less than or equal to $5x10^{-5}$ M, less than or equal to $1x10^{-5}$ M, less than or equal to $5x10^{-6}$ M, less than or equal to $1x10^{-6}$ M, less than or equal to $5x10^{-7}$ M, less than or equal to $1x10^{-7}$ M, less than or equal to $5x10^{-8}$ M, less than or equal to $1x10^{-8}$ M, less than or equal to $5x10^{-9}$ M, less than or equal to $1x10^{-9}$ M, less than or equal to $5x10^{-10}$ M, less than or equal to $1x10^{-10}$ M, less than or equal to $5x10^{-11}$ M, less than or equal to $1x10^{-11}$ M, less than or equal to $5x10^{-12}$ M, or less than or equal to $1x10^{-12}$ M;

13) the binding protein has a higher binding affinity to the peptide-MHC (pMHC) than does a known T-cell receptor, optionally wherein the higher binding affinity is at least 1.05-fold higher;

14) the binding protein induces higher T cell expansion, cytokine release, and/or cytotoxic killing than does a known T-cell receptor when contacted with target cells with a heterozygous expression of MAGEA1, optionally wherein the induction is at least 1.05-fold higher, further optionally wherein the cytotoxic killing is a target cancer cell, optionally wherein the cancer is selected from the group consisting of melanoma, head & neck cancer, lung cancer, cervical cancer, hepatocellular carcinoma, colorectal cancer, gastrointestinal cancer, colorectal cancer, gastrointestinal cancer, breast invasive carcinoma, and bladder urothelial carcinoma; and/or

15) the binding protein does not bind to a peptide-MHC (pMHC) complex comprising a MAGEA10, MAGEA11, RPS6KA2, RPS6KA3, RPS6KA6, STIL, TECPR1, WDR45, CCDC168, SIRPB1, TENM1, ADAMTS20, CPO, SPATA22, and/or ZNF202 peptide epitope;

16) the immunogenic peptide sequence is EADPTGHSY (SEQ ID NO: 14);

17) the binding protein is a TCR; and/or

18) the HLA allele is HLA-A*01:01.

4. An isolated nucleic acid molecule encoding a polypeptide of any one of claims 1-3, optionally wherein the isolated nucleic acid molecule 1) comprises TRAV21 and/or TRAJ47

genes, 2) comprises TRBV5-4 and/or TRBJ2-7 genes, and/or 3) is codon optimized for expression in a host cell.

5. A vector comprising the isolated nucleic acid of claim 4, optionally wherein:

i) the vector is a cloning vector, expression vector, or viral vector;

ii) the vector comprises a vector backbone sequence of any one of SEQ ID NOs: 39-42;

iii) the vector further comprises a nucleic acid sequence encoding CD8α, CD8β, a dominant negative TGFβ receptor II (DN-TGFβRII), selectable protein marker, optionally wherein the selectable protein marker is dihydrofolate reductase (DHFR), optionally wherein the nucleic acid sequence encoding CD8α, CD8β, the DN-TGFβRII, and/or the selectable protein marker is operably linked to a nucleic acid encoding a tag, further optionally wherein the nucleic acid encoding a tag is at the 5' upstream of the nucleic acid sequence encoding CD8α, CD8β, the DN-TGFβRII, and/or the selectable protein marker such that the tag is fused to the N-terminus of CD8α, CD8β, the DN-TGFβRII, and/or the selectable protein marker, further optionally wherein the tag is a CD34 enrichment tag;

iv) the nucleic acid sequence encoding TCRα, TCRβ, CD8α, CD8β, the DN-TGFβRII, and/or the selectable protein marker are interconnected with an internal ribosome entry site or a nucleic acid sequence encoding a self-cleaving peptide, optionally wherein the self-cleaving peptide is P2A, E2A, F2A or T2A;

v) the vector comprises a nucleic acid sequence encoding TCRα, TCRβ, CD8α, CD8β, the DN-TGFβRII, and the selectable protein marker; and/or

vi) the vector comprises a nucleic acid sequence selected from SEQ ID NOs: 39-42.

6. A host cell which comprises the isolated nucleic acid of claim 4, comprises the vector according to claim 5, and/or expresses the binding protein according to any one of claims 1-3, or a population of the host cells, optionally wherein the host cell:

1) is genetically engineered;

2) comprises a chromosomal gene knockout of a TCR gene, an HLA gene, or both;

3) comprises a knockout of an HLA gene selected from an αl macroglobulin gene, α2 macroglobulin gene, α3 macroglobulin gene, β1 microglobulin gene, β2 microglobulin gene, and combinations thereof;

4) comprises a knockout of a TCR gene selected from a TCR α variable region gene, TCR β variable region gene, TCR constant region gene, and combinations thereof;

5) expresses CD8α, CD8β, a DN-TGFβRII, and/or a selectable protein marker, optionally wherein the selectable protein marker is DHFR, further optionally wherein the CD8α, CD8β, the DN-TGFβRII, and/or the selectable protein marker is fused to a CD34 enrichment tag, optionally wherein the host cells are enriched using the CD34 enrichment tag;

6) is a hematopoietic progenitor cell, peripheral blood mononuclear cell (PBMC), cord blood cell, or immune cell;

optionally wherein the immune cell is a T cell, cytotoxic lymphocyte, cytotoxic lymphocyte precursor cell, cytotoxic lymphocyte progenitor cell, cytotoxic lymphocyte stem cell, $CD4^+$ T cell, $CD8^+$ T cell, CD4/CD8 double negative T cell, gamma delta (γδ) T cell, natural killer (NK) cell, NK-T cell, dendritic cell, or a combination thereof;

optionally wherein the T cell:

a) is a naive T cell, central memory T cell, effector memory T cell, or a combination thereof;

b) is a primary T cell or a cell of a T cell line; and/or

c) does not express or has a lower surface expression of an endogenous TCR; and/or

7) the host cell is capable of producing a cytokine or a cytotoxic molecule when contacted with a target cell that comprises a MAGEA1 (EADPTGHSY) peptide-MHC (HLA-A*01) (pMHC) complex,

optionally wherein

a) the host cell is contacted with the target cell *in vitro, ex vivo,* or *in vivo*;

b) the cytokine is TNF-α, IL-2, and/or IFN-γ;

c) the cytotoxic molecule is perforins and/or granzymes, optionally wherein the cytotoxic molecule is granzyme B;

d) the host cell is capable of producing a higher level of cytokine or a cytotoxic molecule when contacted with a target cell with a heterozygous expression of MAGEA1,

optionally wherein the host cell is capable of producing an at least 1.05-fold higher level of cytokine or a cytotoxic molecule;

e) the host cell is capable of killing a target cell that comprises a MAGEA1 (EADPTGHSY) peptide-MHC (HLA-A*01) (pMHC) complex, optionally wherein

i) the killing is determined by a killing assay;

ii) the ratio of the host cell and the target cell in the killing assay is from 20:1 to 1:4;

iii) the target cell is a target cell pulsed with 1 μg/mL to 50 pg/mL of MAGEA1 peptide, optionally wherein the target cell is a cell monoallelic for an MHC matched to the MAGEA1 peptide;

iv) the host cell is capable of killing a higher number of target cells when contacted with target cells with a heterozygous expression of MAGEA1, optionally wherein the cell killing is at least 1.05-fold higher; and/or

v) the target cell is cell line or a primary cell, optionally wherein the target cell is selected from the group consisting of a HEK293 derived cell line, a cancer cell line, a primary cancer cell, a transformed cell line, and an immortalized cell line;

f) the MHC is an MHC multimer, optionally wherein the MHC multimer is a tetramer;

g) the MHC alpha chain is encoded by an HLA-A*01:01 allele;

h)

i) the host cell does not induce T cell expansion, cytokine release, or cytotoxic killing when contact with a target cell that comprises a peptide-MHC (pMHC) complex comprising a MAGEA10, MAGEA11, RPS6KA2, RPS6KA3, RPS6KA6, STIL, TECPR1, WDR45, CCDC168, SIRPB1, TENM1, ADAMTS20, CPO, SPATA22, and/or ZNF202 peptide epitope, and/or

ii) the host cell does not express MAGEA1 antigen, is not recognized by a binding protein of any one of claims 1-3, is not of serotype HLA-A*01, and/or does not express an HLA-A*01 allele; and/or

i) the HLA allele is HLA-A*01:01.

7. A composition comprising a) a binding protein according to any one of claims 1-3, b) an isolated nucleic acid according to claim 4, c) a vector according to claim 5, d) a host cell according to claim 6, and/or e) a population of host cells according to claim 6, and a carrier.

8. A device or kit comprising a) a binding protein according to any one of claims 1-3, b) an isolated nucleic acid according to claim 4, c) a vector according to any one of claim 5, d) a host cell according to claim 6, and/or e) a population of host cells according to claim 6, said device or kit optionally comprising a reagent to detect binding of a), d) and/or e) to a pMHC complex.

9. A method of producing a binding protein according to any one of claims 1-3, wherein the method comprises the steps of: (i) culturing a transformed host cell which has been transformed by a nucleic acid comprising a sequence encoding a binding protein according to any one of claims 1-3 under conditions suitable to allow expression of said binding protein; and (ii) recovering the expressed binding protein.

10. A method of producing a host cell expressing a binding protein according to any one of claims 1-3, wherein the method comprises the steps of: (i) introducing a nucleic acid comprising a sequence encoding a binding protein according to any one of claims 1-3 into the host cell; and (ii) culturing the transformed host cell under conditions suitable to allow expression of said binding protein.

11. A method of detecting the presence or absence of a MAGEA1 (EADPTGHSY) peptide-MHC (HLA-A*01) (pMHC) complex and/or a cell expressing the pMHC complex, optionally wherein the cell is a hyperproliferative cell, comprising detecting the presence or absence of said pMHC complex in a sample by use of at least one binding protein according to any one of claims 1-3, at least one host cell according to claim 6, or a population of host cells according to claim 6, wherein detection of the pMHC complex is indicative of the presence of the pMHC complex and/or cell expressing the pMHC complex;

optionally wherein:

1) the pMHC complex is detected in the form of fluorescence activated cell sorting (FACS), enzyme linked immunosorbent assay (ELISA), radioimmune assay (RIA), immunochemically, Western blot, or intracellular flow assay;

2) the method further comprises obtaining the sample from a subject;

3) the level of reactivity is indicated by

A) the presence of binding and/or

B) T cell activation and/or effector function, optionally wherein

i) the T cell activation or effector function is T cell proliferation, killing, or cytokine release; and/or

ii) the T cell binding, activation, and/or effector function is detected using fluorescence activated cell sorting (FACS), enzyme linked immunosorbent assay (ELISA), radioimmune assay (RIA), immunochemically, Western blot, or intracellular flow assay; and/or

4) the alpha chain of the MHC is encoded by an HLA-A*01:01 allele.

12. A method of detecting the level of a disorder characterized by MAGEA1 expression in a subject expressing HLA-A*01 serotype, comprising:

a) contacting a sample obtained from the subject with at least one binding protein according to any one of claims 1-3, at least one host cell according to claim 6, or a population of host cells according to claim 6; and

b) detecting the level of reactivity,

wherein the presence or a higher level of reactivity compared to a control level indicates the level of the disorder characterized by MAGEA1 expression in the subject;

optionally wherein:

i) the control level is:

A) a reference number, or

B) a level from a subject without the disorder characterized by MAGEA1 expression;

ii) the level of reactivity is indicated by

A) the presence of binding and/or

B) T cell activation and/or effector function, optionally wherein

1) the T cell activation or effector function is T cell proliferation, killing, or cytokine release; and/or

2) the T cell binding, activation, and/or effector function is detected using fluorescence activated cell sorting (FACS), enzyme linked immunosorbent assay (ELISA), radioimmune assay (RIA), immunochemically, Western blot, or intracellular flow assay; and/or

iii) the alpha chain of the HLA-A*01 serotype MHC expressed by the subject is encoded by an HLA-A*01:01 allele.

13. A method for monitoring the progression of a disorder characterized by MAGEA1 expression in a subject expressing HLA-A*01 serotype, the method comprising:

a) detecting in a subject sample the presence or level of reactivity between a sample obtained from the subject and at least one binding protein according to any one of claims 1-3, at least one host cell according to claim 6, or a population of host cells according to claim 6;

b) repeating step a) at a subsequent point in time; and

c) comparing the level of MAGEA1 or the cell of interest expressing MAGEA1 detected in steps a) and b) to monitor the progression of the disorder characterized by MAGEA1 expression in the subject, wherein an absent or reduced MAGEA1 level or the cell of interest expressing MAGEA1 detected in step b) compared to step a) indicates an inhibited progression of the disorder characterized by MAGEA1 expression in the subject and a presence or increased MAGEA1 level or the cell of interest expressing MAGEA1 detected in step b) compared to step a) indicates a progression of the disorder characterized by MAGEA1 expression in the subject,

optionally wherein:

i) between the first point in time and the subsequent point in time, the subject has undergone treatment to treat the disorder characterized by MAGEA1 expression;

ii) the level of reactivity is indicated by

A) the presence of binding and/or

B) T cell activation and/or effector function, optionally wherein

1) the T cell activation or effector function is T cell proliferation, killing, or cytokine release; and/or

2) the T cell binding, activation, and/or effector function is detected using fluorescence activated cell sorting (FACS), enzyme linked immunosorbent assay (ELISA), radioimmune assay (RIA), immunochemically, Western blot, or intracellular flow assay; and/or

iii) the alpha chain of the HLA-A*01 serotype MHC expressed by the subject is encoded by an HLA-A*01:01 allele.

14. A method for predicting the clinical outcome of a subject expressing HLA-A*01 serotype and afflicted with a disorder characterized by MAGEA1 expression comprising:

a) determining the presence or level of reactivity between a sample obtained from the subject and at least one binding protein according to any one of claims 1-3, at least one host cell according to claim 6, or a population of host cells according to claim 6; and

b) comparing the presence or level of reactivity to that from a control, wherein the control is obtained from a subject having a good clinical outcome;

wherein the absence or a reduced level of reactivity in the subject sample as compared to the control indicates that the subject has a good clinical outcome, optionally wherein:

i) the level of reactivity is indicated by

A) the presence of binding; and/or

B) T cell activation and/or effector function, optionally wherein

1) the T cell activation or effector function is T cell proliferation, killing, or cytokine release; and/or

2) the T cell binding, activation, and/or effector function is detected using fluorescence activated cell sorting (FACS), enzyme linked immunosorbent assay (ELISA), radioimmune assay (RIA), immunochemically, Western blot, or intracellular flow assay; and/or

ii) the alpha chain of the HLA-A*01 serotype MHC expressed by the subject is encoded by an HLA-A*01:01 allele.

15. A method of assessing the efficacy of a therapy for a disorder characterized by MAGEA1 expression in a subject expressing HLA-A*01 serotype comprising:

a) determining the presence or level of reactivity between a sample obtained from the subject and at least one binding protein according to any one of claims 1-3, at least one host cell

according to claim 6, or a population of host cells according to claim 6, in a first sample obtained from the subject prior to providing at least a portion of the therapy for the disorder characterized by MAGEA1 expression to the subject, and

b) determining the presence or level of reactivity between a sample obtained from the subject and at least one binding protein according to any one of claims 1-3, at least one host cell according to claim 6, or a population of host cells according to claim 6, in a second sample obtained from the subject following provision of the therapy for the disorder characterized by MAGEA1 expression,

wherein the absence or a reduced level of reactivity in the second sample, relative to the first sample, is an indication that the therapy is efficacious for treating the disorder characterized by MAGEA1 expression in the subject, and wherein the presence or an increased level of reactivity in the second sample, relative to the first sample, is an indication that the therapy is not efficacious for treating the disorder characterized by MAGEA1 expression in the subject, optionally wherein:

i) the level of reactivity is indicated by

A) the presence of binding; and/or

B) T cell activation and/or effector function, optionally wherein

1) the T cell activation or effector function is T cell proliferation, killing, or cytokine release; and/or

2) the T cell binding, activation, and/or effector function is detected using fluorescence activated cell sorting (FACS), enzyme linked immunosorbent assay (ELISA), radioimmune assay (RIA), immunochemically, Western blot, or intracellular flow assay; and/or

ii) the alpha chain of the HLA-A*01 serotype MHC expressed by the subject is encoded by an HLA-A*01:01 allele.

16. A composition comprising an agent selected from the group consisting of a) a binding protein of any one of claims 1-3, b) an isolated nucleic acid according to claim 4, c) a vector according to claim 5, and d) a host cell or a population of host cells according to claim 6 for use in preventing and/or treating a disorder characterized by MAGEA1 expression in a subject

expressing HLA-A*01 serotype, wherein the composition is adapted to be administrable to the subject expressing MAGEA1 in a therapeutically effective amount, optionally wherein:

1) the composition is adapted to be administrable to a subject;

2) the cell is an allogeneic cell, syngeneic cell, or autologous cell;

3) the cell is a host cell according to claim 6 or a population of the host cells;

4) the target cell is a cancer cell expressing MAGEA1;

5) the composition further comprises a pharmaceutically acceptable carrier;

6) the composition induces an immune response against the target cell expressing MAGEA1 in the subject, optionally wherein the composition induces an antigen-specific T cell immune response against the target cell expressing MAGEA1 in the subject, optionally wherein the antigen-specific T cell immune response comprises at least one of a $CD4^+$ helper T lymphocyte (Th) response and a CD8+ cytotoxic T lymphocyte (CTL) response;

7) the use further comprises at least one additional treatment for the disorder characterized by MAGEA1 expression adapted to be administrable, optionally wherein the at least one additional treatment for the disorder characterized by MAGEA1 expression is adapted to be administrable concurrently or sequentially with the composition;

8) the disorder characterized by MAGEA1 expression is a cancer or relapse thereof, optionally wherein the cancer is selected from the group consisting of melanoma, head & neck cancer, lung cancer, cervical cancer, hepatocellular carcinoma, breast invasive carcinoma, and bladder urothelial carcinoma;

9) the subject is an animal model of a disorder characterized by MAGEA1 expression and/or a mammal, optionally wherein the mammal is a human, a primate, or a rodent; and/or

10) the alpha chain of the HLA-A*01 serotype MHC expressed by the subject is encoded by an HLA-A*01:01 allele.

17. Use of a composition comprising an agent selected from the group consisting of a) a binding protein of any one of claims 1-3, b) an isolated nucleic acid according to claim 4, c) a vector according to claim 5, and d) a host cell or population of host cells according to claim 6 in the manufacture of a medicament for preventing and/or treating a disorder characterized by MAGEA1 expression in a subject expressing HLA-A*01 serotype, wherein the composition is

adapted to be administrable to the subject expressing MAGEA1 in a therapeutically effective amount, optionally wherein:

1) the composition is adapted to be administrable to a subject;

2) the cell is an allogeneic cell, syngeneic cell, or autologous cell;

3) the cell is a host cell according to claim 6 or a population of the host cells;

4) the target cell is a cancer cell expressing MAGEA1;

5) the composition further comprises a pharmaceutically acceptable carrier;

6) the composition induces an immune response against the target cell expressing MAGEA1 in the subject, optionally wherein the composition induces an antigen-specific T cell immune response against the target cell expressing MAGEA1 in the subject, optionally wherein the antigen-specific T cell immune response comprises at least one of a $CD4^+$ helper T lymphocyte (Th) response and a CD8+ cytotoxic T lymphocyte (CTL) response;

7) the use further comprises at least one additional treatment for the disorder characterized by MAGEA1 expression adapted to be administrable, optionally wherein the at least one additional treatment for the disorder characterized by MAGEA1 expression is adapted to be administrable concurrently or sequentially with the composition;

8) the disorder characterized by MAGEA1 expression is a cancer or relapse thereof, optionally wherein the cancer is selected from the group consisting of melanoma, head & neck cancer, lung cancer, cervical cancer, hepatocellular carcinoma, breast invasive carcinoma, and bladder urothelial carcinoma;

9) the subject is an animal model of a disorder characterized by MAGEA1 expression and/or a mammal, optionally wherein the mammal is a human, a primate, or a rodent; and/or

10) the alpha chain of the HLA-A*01 serotype MHC expressed by the subject is encoded by an HLA-A*01:01 allele.