

CLAIMS

What is claimed is:

1. A polynucleotide that encodes an anti-CD70 chimeric antigen receptor (CAR) comprising a signal peptide, an anti-CD70 antigen binding domain, transmembrane domain, and at least one intracellular domain, wherein the anti-CD70 antigen binding domain does not comprise an antibody, and optionally wherein a transmembrane domain of the CAR is not a CD27 transmembrane domain.
2. The polynucleotide of claim 1, wherein the anti-CD70 antigen binding domain is comprised in an extracellular domain of CD27, optionally, wherein the sequence that encodes the antigen binding domain is codon optimized.
3. The polynucleotide of claim 1 or 2, wherein the antigen binding domain comprises, consists of, or consists essentially of SEQ ID NO:2.
4. The polynucleotide of any one of claims 1-3, wherein the transmembrane domain is from CD28, the alpha chain of the T- cell receptor, beta chain of the T- cell receptor, zeta chain of the T- cell receptor, CD3 zeta, CD3 epsilon, CD3 gamma, CD3 delta, CD45, CD4, CD5, CD8, CD9, CD 16, CD22, CD33, CD37, CD64, CD80, CD86, CD 134, CD137, CD154, ICOS/CD278, GITR/CD357, NKG2D, DAP10, or DAP12, optionally, wherein the sequence that encodes the transmembrane domain is codon optimized.
5. The polynucleotide of any one of claims 1-4, wherein the transmembrane domain comprises, consists of, or consists essentially of SEQ ID NO:3 or SEQ ID NO:7.
6. The polynucleotide of any one of claims 1-5, wherein the CAR comprises two, three, or more intracellular domains, optionally, wherein:
 - the intracellular domain comprises CD3zeta;
 - the intracellular domain comprises an ITAM-containing signaling domain; and/or
 - the intracellular domain is an intracellular costimulatory domain, wherein the intracellular costimulatory domain is selected from the group consisting of CD27, CD28, 4-

1BB, DAP12, NKG2D, OX-40 (CD134), DAP10, CD40L, 2B4, DNAM, CS1, CD48, NKp30, NKp44, NKp46, NKp80, and a combination thereof.

7. The polynucleotide of any one of claims 1-6, wherein the intracellular domain comprises one or more of SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:11, or SEQ ID NO:12.

8. The polynucleotide of any one of claims 1-7, wherein the signal peptide is from CD27 or granulocyte-macrophage colony-stimulating factor receptor (GMSCF-R), optionally, wherein the sequence of the CD27 signal peptide comprises, consists of, or consists essentially of SEQ ID NO:6.

9. The polynucleotide of any one of claims 1-8, wherein the signal peptide is from CD27, the anti-CD70 antigen binding domain is from CD27, the transmembrane domain is from CD27, and an intracellular domain is from CD27.

10. The polynucleotide of any one of claims 1-9, wherein the CAR comprises, consists of, or consists essentially of SEQ ID NO:1, SEQ ID NO:4, or SEQ ID NO:5.

11. The polynucleotide of any one of claims 1-10, wherein the CAR comprises one or more of the following:

- (a) CD27 signal peptide (SP), CD27 extracellular domain (EC), CD27 transmembrane domain (TMD), DAP12 intracellular domain (ICD), and CD3zeta;
- (b) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD27 TMD, DAP12 ICD;
- (c) CD27 SP, CD27 EC, CD28 TMD, DAP12 ICD, and CD3zeta;
- (d) GMSCF-R SP, codon optimized CD27 EC, CD28 TMD, DAP12 ICD, and CD3zeta;
- (e) CD27 SP, CD27 EC, CD27 TMD, Natural killer group 2 member D (NKG2D) ICD, and CD3zeta;
- (f) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD27 TMD, NKG2D ICD, and CD3zeta;

- (g) CD27 SP, CD27 EC, CD28 TMD, Natural killer group 2 member D (NKG2D) ICD, and CD3zeta;
- (h) GMSCF-R SP, codon optimized CD27 EC, CD28 TMD, NKG2D ICD, and CD3zeta;
- (i) CD27 SP, CD27 EC, CD27 TMD, 4-1BB ICD, and CD3zeta;
- (j) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD27 TMD, 4-1BB ICD, and CD3zeta;
- (k) CD27 SP, CD27 EC, CD28 TMD, 4-1BB ICD, and CD3zeta;
- (l) GMSCF-R SP, codon optimized CD27 EC, CD28 TMD, 4-1BB ICD, and CD3zeta;
- (m) CD27 SP, CD27 EC, CD27 TMD, DAP10 ICD, and CD3zeta;
- (n) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD27 TMD, DAP10 ICD, and CD3zeta;
- (o) CD27 SP, CD27 EC, CD28 TMD, DAP10 ICD, and CD3zeta;
- (p) GMSCF-R SP, codon optimized CD27 EC, CD28 TMD, DAP10 ICD, and CD3zeta;
- (q) CD27 SP, CD27 full length (FL), CD27 TMD, CD27 ICD, and CD3zeta;
- (r) GMSCF-R SP, codon optimized CD27 FL, codon optimized CD27 TMD, codon optimized CD27 ICD, and CD3zeta;
- (s) CD27 SP, CD27 FL, CD27 TMD, CD27 ICD, CD28 ICD, and CD3zeta;
- (t) GMSCF-R SP, codon optimized CD27 FL, codon optimized CD27 TMD, codon optimized CD27 ICD, CD28 ICD, and CD3zeta;
- (u) CD27 SP, CD27 FL, CD27 TMD, CD27 ICD, 4-1BB ICD, and CD3zeta;
- (v) GMSCF-R SP, codon optimized CD27 FL, codon optimized CD27 TMD, codon optimized CD27 ICD, 4-1BB ICD, and CD3zeta;
- (w) CD27 SP, CD27 FL, CD27 TMD, CD27 ICD, DAP10 ICD, and CD3zeta;
- (x) GMSCF-R SP, CD27 FL, CD27 TMD, CD27 ICD, DAP10 ICD, and CD3zeta;
- (y) CD27 SP, CD27 FL, CD27 TMD, CD27 ICD, DAP12 ICD, and CD3zeta;
- (z) GMSCF-R SP, codon optimized CD27 FL, codon optimized CD27 TMD, codon optimized CD27 ICD, DAP12 ICD, and CD3zeta;
- (aa) CD27 SP, CD27 FL, CD27 TMD, CD27 ICD, NKG2D ICD, and CD3zeta;

- (bb) GMSCF-R SP, codon optimized CD27 FL, codon optimized CD27 TMD, codon optimized CD27 ICD, NKG2D ICD, and CD3zeta;
 - (cc) CD27 SP, CD27 EC, CD27 TMD, and CD3zeta;
 - (dd) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD27 TMD, and CD3zeta;
 - (ee) CD27 SP, CD27 EC, CD28 TMD, and CD3zeta;
 - (ff) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD28 TMD, and CD3zeta;
 - (gg) CD27 SP, CD27 EC, CD27 TMD, CD28 ICD, and CD3zeta;
 - (hh) GMSCF-R SP, codon optimized CD27 EC, codon optimized CD27 TMD, CD28 ICD, and CD3zeta;
 - (ii) CD27 SP, CD27 EC, CD28 TMD, CD28 ICD, and CD3zeta; or
 - (jj) GMSCF-R SP, codon optimized CD27 EC, CD28 TMD, and CD3zeta.
12. An immune cell comprising the polynucleotide of any one of claims 1-11.
 13. A population of the immune cells of claim 12, said cells present in a suitable medium.
 14. Use of the polynucleotide of any one of claims 1-11 in the manufacture of a medicament for killing CD70-positive cells in an individual, wherein a therapeutically effective amount of cells harboring the polynucleotide of any one of claims 1-11 is to be administered to the individual.
 15. The use of claim 14, further comprising an effective amount of an additional therapy to be provided to the individual and/or identifying CD70-positive cells in the individual.
 16. The polynucleotide of any one of claims 1-11 for use in therapy.