

CLAIMS

What is claimed is:

1. A genetically modified anti-B7-H4 CAR NK cell, comprising:
 - a recombinantly expressed cytokine, wherein the recombinantly expressed cytokine ~~comprises-is~~ IL-2 or IL-15;
 - a recombinantly expressed CD16;
 - a membrane bound recombinantly expressed anti-B7-H4 chimeric antigen receptor (CAR) that comprises in a single polypeptide chain an extracellular binding domain, a hinge domain, a transmembrane domain, and ~~an~~ FcεRIγ signaling domain ~~and/or a CD3zeta signaling domain~~;wherein the recombinantly expressed cytokine, the recombinantly expressed CD16, and the anti-B7-H4 CAR are expressed from an expression cassette of a plasmid that is transfected into the NK cell; and wherein the NK cell is an NK-92 cell.
2. The genetically modified NK cell of claim 1 wherein the recombinantly expressed cytokine comprises an endoplasmic retention sequence.
3. The genetically modified NK cell of claim 1 wherein the recombinantly expressed CD16 is a high-affinity CD16 variant having a 158V mutation.
4. The genetically modified NK cell of claim 1 wherein the extracellular binding domain comprises a scFv.
5. The genetically modified NK cell of claim 1 wherein the anti-B7-H4 CAR has an amino acid sequence of SEQ ID NO:1.
6. The genetically modified NK cell of claim 1 wherein recombinantly expressed cytokine, the recombinantly expressed CD16, and the recombinantly expressed CAR are expressed from a tricistronic recombinant nucleic acid.
7. The genetically modified NK cell of claim 1 wherein the recombinantly expressed cytokine and/or the recombinantly expressed CD16 is expressed from a recombinant nucleic acid that is integrated into the genome of the NK cell.

8. A recombinant nucleic acid plasmid, comprising:
 - a first sequence portion encoding a cytokine, wherein the cytokine ~~comprises~~is IL-2 or IL-15;
 - a second sequence portion encoding a CD16;
 - a third sequence portion encoding an anti-B7-H4 CAR that comprises in a single polypeptide chain an extracellular binding domain, a hinge domain, a transmembrane domain, and an FcεRIγ signaling domain~~and/or a CD3zeta signaling domain~~;wherein the first, second, and third sequence portions encoding the cytokine, the recombinantly expressed CD16, and the anti-B7-H4 CAR, respectively, form an expression cassette on the plasmid.
9. The recombinant nucleic acid of claim 8 wherein the expression cassette is a tricistronic DNA.
10. The recombinant nucleic acid of claim 8 wherein the cytokine is IL-2.
11. The recombinant nucleic acid of claim 8 wherein the cytokine comprises an endoplasmic retention sequence.
12. The recombinant nucleic acid of claim 8 wherein the CD16 is a high-affinity CD16 variant having a 158V mutation.
13. The recombinant nucleic acid of claim 8 wherein the extracellular binding domain comprises a scFv.
14. The recombinant nucleic acid of claim 8 wherein the hinge domain and/or the transmembrane domain comprise a CD8 hinge domain and/or a CD28 transmembrane domain.
15. The recombinant nucleic acid of claim 8 wherein the anti-B7-H4 CAR has an amino acid sequence of SEQ ID NO:1.
16. A recombinant cell comprising the recombinant nucleic acid of any one of claims 8-15.
17. The recombinant cell of claim 16 wherein the cell is a bacterial cell.
18. The recombinant cell of claim 16 wherein the cell is an autologous NK cell.

19. The recombinant cell of claim 18 wherein the NK cell is an NK-92 cell that is optionally genetically modified.
20. Use of a therapeutically effective amount of the genetically modified NK cell of any one of claims 1-7 in the manufacture of a medicament for the treatment of cancer in a patient, wherein the medicament is to be administered to the patient.
21. The use of claim 20, wherein the medicament is to be administered in combination with at least one additional therapeutic entity selected from the group consisting of a viral cancer vaccine, a bacterial cancer vaccine, a yeast cancer vaccine, N-803, a bi-specific engager, an antibody, a stem cell transplant, a primary NK cell, and a tumor targeted cytokine.
22. The use of claim 20 or 21, wherein the cancer is selected from leukemia, acute lymphocytic leukemia, acute myelocytic leukemia, chronic leukemias, chronic myelocytic (granulocytic) leukemia, chronic lymphocytic leukemia, polycythemia vera, lymphomas, Hodgkin's disease, non-Hodgkin's disease, multiple myeloma, Waldenstrom's macroglobulinemia, heavy chain disease, solid tumors including, ~~but not limited to,~~ sarcomas and carcinomas such as fibrosarcoma, myxosarcoma, liposarcoma, chondrosarcoma, osteogenic sarcoma, chordoma, angiosarcoma, endotheliosarcoma, lymphangiosarcoma, lymphangioendotheliosarcoma, synovioma, mesothelioma, Ewing's tumor, leiomyosarcoma, rhabdomyo sarcoma, colon carcinoma, pancreatic cancer, breast cancer, ovarian cancer, prostate cancer, squamous cell carcinoma, basal cell carcinoma, adenocarcinoma, sweat gland carcinoma, sebaceous gland carcinoma, papillary carcinoma, papillary adenocarcinomas, cystadenocarcinoma, medullary carcinoma, bronchogenic carcinoma, renal cell carcinoma, hepatoma, bile duct carcinoma, choriocarcinoma, seminoma, embryonal carcinoma, Wilm's tumor, cervical cancer, testicular tumor, lung carcinoma, small cell lung carcinoma, bladder carcinoma, epithelial carcinoma, glioma, astrocytoma, medulloblastoma, craniopharyngioma, ependymoma, pinealoma, hemangioblastoma, acoustic neuroma, oligodendroglioma, menangioma, melanoma, neuroblastoma and retinoblastoma.
23. The genetically modified NK cell of any one of claims 1-7 for use as a medicament.