

What is claimed is:

1. A method of genetically modifying an NK cell comprising
  - a) obtaining guide RNA (gRNA) specific for the target DNA sequence; and
  - b) introducing via electroporation into a target NK cell, a ribonucleoprotein (RNP)
- 5 complex comprising a class 2 CRISPR/Cas endonuclease (Cas9) complexed with a corresponding CRISPR/Cas guide RNA that hybridizes to a target sequence within the genomic DNA of the NK cell.
2. The method of claim 1, wherein the genome of the NK cell is modified by insertion or deletion of one or more base pairs, by insertion of a heterologous DNA fragment (e.g., the donor polynucleotide), by deletion of an endogenous DNA fragment, by inversion or translocation of an  
10 endogenous DNA fragment, or a combination thereof.
3. The method of claim 1, wherein the NK cells are primary or expanded NK cells.
4. The method of claim 3, wherein the primary NK cells are incubated for 2, 3, or 4 days in the presence of IL-2 prior to electroporation.
5. The method of claim 3, wherein the primary NK cells are expanded for 4 days in the presence  
15 of irradiated feeder cells prior to electroporation.
6. The method of claim 1, further comprising expanding the modified NK cells with irradiated mBIL-21 expressing feeder cells following electroporation.
7. The method of claim 1, wherein the method further comprises forming the RNP complex by diluting 36  $\mu$ M cas9 into a solution of 200  $\mu$ M crRNA and TracerRNA.
- 20 8. An NK cell modified by the method of claim 1.
9. A genetically modified NK cell comprising a knockout of the gene encoding the transforming growth factor- $\beta$  receptor 2 (TGFB2) or hypoxanthine phosphoribosyltransferase 1 (HPRT1).
10. Use of an engineered NK cell of claim 8 in the manufacture of a medicament for the treatment of a disease in a subject.
- 25 11. The use of claim 10, wherein the subject has a cancer.
12. The use of claim 10, wherein the NK cell is a primary NK cell that has been modified ex vivo and after modification is to be transferred to the subject.
13. The use of claim 10, wherein the NK cell is an autologous NK cell.

14. The use of claim 10, wherein the NK cell is from an allogeneic donor source.
15. The use of claim 10, wherein the NK cell is expanded with irradiated mblL-21 expressing feeder cells prior to administration to the subject.
16. The use of claim 10, wherein the RNP complex targets the TGFRB2 or HPRT1 gene.
- 5 17. The use of claim 10, wherein the NK cell has been modified to comprise a knockout of the TGFRB2 gene.