WHAT IS CLAIMED IS:

1. A recombinant polypeptide comprising:

a protein of interest, wherein the protein of interest is a chimeric antigen receptor (CAR) or a T cell receptor (TCR);

a protein localization tag; and

a protease cleavage site disposed between the protein of interest and the protein localization tag.

2. The recombinant polypeptide of claim 1, comprising from N-terminus to C-terminus:

(a) the protein of interest; the protease cleavage site; and the protein localization tag; or

(b) the protein localization tag, the protease cleavage site; and the protein of interest

.

~~3. The recombinant polypeptide of claim 1, comprising from N-terminus to C-terminus:~~

~~the protein localization tag,~~

~~the protease cleavage site; and~~

~~the protein of interest.~~

~~4.~~3. The recombinant polypeptide of any one of claims 1 to 2~~3~~, wherein the protein localization tag is selected from the group consisting of: an endoplasmic reticulum (ER) localization tag, a Golgi apparatus (Golgi) localization tag, a lysosome localization tag, a plasma membrane localization tag, a mitochondria localization tag, a peroxisome localization tag, a cytosolic localization tag, and a nuclear localization tag.

~~5.~~4. The recombinant polypeptide of any one of claims 1 to 3~~4~~, wherein the protein localization tag is an ER localization tag.

~~6.~~5. The recombinant polypeptide of claim 4~~5~~, wherein the ER localization tag comprises 85% or greater, 90% or greater, or 100% amino acid sequence identity to the amino acid sequence LYKYKSRRSFIDEKKMP (SEQ ID NO:1) or 100% amino acid sequence identity to the amino acid sequence KKMP (SEQ ID NO:2).

~~7. The recombinant polypeptide of claim 5, wherein the ER localization tag comprises the amino acid sequence KKMP (SEQ ID NO:2).~~

~~8.~~6. The recombinant polypeptide of any one of claims 1 to 3~~4~~, wherein the protein localization tag is a Golgi localization tag, wherein the Golgi localization tag comprises the amino acid sequence YQRL (SEQ ID NO:24).

~~9. The recombinant polypeptide of claim 8, wherein the Golgi localization tag comprises the amino acid sequence YQRL (SEQ ID NO:24).~~

~~10.~~7. The recombinant polypeptide of any one of claims 1 to 3~~4~~, wherein the protein localization tag is a lysosome localization tag, wherein the lysosome localization tag comprises the amino acid sequence KFERQ (SEQ ID NO:25).

~~11. The recombinant polypeptide of claim 10, wherein the lysosome localization tag comprises the amino acid sequence KFERQ (SEQ ID NO:25).~~

~~12. The recombinant polypeptide of any one of claims 1 to 11, wherein the protein of interest is not engineered.~~

~~13. The recombinant polypeptide of any one of claims 1 to 11, wherein the protein of interest is engineered.~~

~~14. The recombinant polypeptide of claim 13, wherein the engineered protein of interest is an engineered cell surface receptor.~~

~~15. The recombinant polypeptide of claim 14, wherein the engineered cell surface receptor is a chimeric antigen receptor (CAR).~~

~~16.~~8. The recombinant polypeptide of any one of claims 1 to 7~~claim 15~~, wherein the CAR comprises a first and a second intracellular signaling domain or fragments thereof independently selected from the group consisting of: a CD3 intracellular signaling domain, a CD28 intracellular signaling domain, a 41 BB intracellular signaling domain, an OX40 intracellular signaling domain, an inducible co-stimulator (ICOS) intracellular signaling domain, an CD27 intracellular signaling domain, and a MyD88/CD40 intracellular signaling domain.

~~17.~~9. The recombinant polypeptide of claim 8~~16~~, wherein the first intracellular signaling domain or fragment thereof is a CD3 intracellular signaling domain and the second intracellular signaling domain or fragment thereof is a 4-1BB intracellular signaling domain or a CD28 intracellular signaling domain.

~~18. The recombinant polypeptide of claim 16, wherein the first intracellular signaling domain or fragment thereof is a CD3 intracellular signaling domain and the second intracellular signaling domain or fragment thereof is a CD28 intracellular signaling domain.~~

~~19.~~10. The recombinant polypeptide of any one of claims 1~~5~~ to 9~~18~~, wherein the CAR comprises an extracellular binding domain comprising~~of the CAR comprises~~ a single chain antibody or a single chain variable fragment (scFv).

~~20. The recombinant polypeptide of claim 19, wherein the single chain antibody is a single chain variable fragment (scFv).~~

~~21.~~11. The recombinant polypeptide of claim 10~~9 or claim 20~~, wherein the extracellular binding domain of the CAR specifically binds an antigen expressed on the surface of a cancer cell, wherein the cancer cell-surface antigen is selected from the group consisting of: B7H3 (CD276), CD19, GD2, CD22, and HER2.

~~22. The recombinant polypeptide of claim 21, wherein the cancer cell-surface antigen is selected from the group consisting of: B7H3 (CD276), CD19, GD2, CD22, and HER2.~~

~~23. The recombinant polypeptide of claim 13, wherein the engineered protein of interest is a T cell receptor (TCR).~~

~~24. The recombinant polypeptide of claim 23, wherein the TCR recognizes an antigen complexed with a major histocompatibility complex (MHC) molecule displayed on the surface of a cancer cell.~~

~~25. The recombinant polypeptide of any one of claims 1 to 13, wherein the protein of interest is a cell surface molecule.~~

~~26. The recombinant polypeptide of claim 25, wherein the cell surface molecule is a cell surface receptor.~~

~~27. The recombinant polypeptide of claim 25 or claim 26, wherein the cell surface molecule is selected from the group consisting of: a cytokine receptor, a chemokine receptor, an adhesion molecule, an integrin, an inhibitory receptor, an inhibitory cell surface ligand, a stimulatory receptor, a stimulatory cell surface ligand, an immunoreceptor tyrosine-based activation motif (ITAM)-containing receptor, and an immunoreceptor tyrosine-based inhibition motif (ITIM)-containing receptor.~~

~~28. The recombinant polypeptide of any one of claims 1 to 13, wherein the protein of interest is a transcription factor.~~

~~29. The recombinant polypeptide of any one of claims 1 to 13, wherein the protein of interest is a secreted effector molecule.~~

~~30. The recombinant polypeptide of claim 29, wherein the secreted effector molecule is selected from the group consisting of: a stimulatory ligand, an inhibitory ligand, a cytokine, a chemokine, a growth factor, and a protease.~~

~~31. The recombinant polypeptide of claim 29 or claim 30, wherein the protein localization tag is an ER localization tag.~~

~~32.~~12. The recombinant polypeptide of any one of claims 1 to ~~3~~11, wherein the protease cleavage site is a viral protease cleavage site.

~~33.~~13. The recombinant polypeptide of claim ~~3~~12, wherein the viral protease cleavage site is for a viral protease derived from hepatitis C virus (HCV) nonstructural protein 3 (NS3).

~~34.~~14. The recombinant polypeptide of claim ~~3~~13, wherein the viral protease cleavage site is for a viral protease that further comprises a cofactor polypeptide derived from HCV nonstructural protein 4A (NS4A).

~~35.~~15. The recombinant polypeptide of any one of claims ~~3~~12 to ~~3~~14, wherein the viral protease cleavage site is selected from the group consisting of: an NS4A/4B junction cleavage site, an NS3/NS4A junction cleavage site, an NS4A/NS4B junction cleavage site, an NS4B/NS5A junction cleavage site, an NS5A/NS5B junction cleavage site, and variants thereof cleavable by the viral protease.

~~36. The recombinant polypeptide of any one of claims 1 to 35, further comprising a reporter domain.~~

~~37. The recombinant polypeptide of claim 36, wherein the reporter domain comprises a fluorescent protein.~~

~~38. The recombinant polypeptide of claim 37, wherein the fluorescent protein is green fluorescent protein.~~

~~39. The recombinant polypeptide of claim 36, wherein the reporter domain comprises a bioluminescent protein.~~

~~40. The recombinant polypeptide of claim 39, wherein the bioluminescent protein is a luciferase.~~

~~41. The recombinant polypeptide of claim 40, wherein the luciferase is a nanoluciferase.~~

~~42. The recombinant polypeptide of any one of claims 36 to 41, wherein the reporter domain is disposed between the protein of interest and the protease cleavage site.~~

~~43.~~16. The recombinant polypeptide of any one of claims 1 to 15~~42~~, wherein the recombinant polypeptide further comprises a protease, and wherein the protease cleavage site is a cleavage site for the protease, wherein the protease cleavage site is disposed between the protein of interest and the protease.

~~44. The recombinant polypeptide of claim 43, wherein the protease cleavage site is disposed between the protein of interest and the protease.~~

~~45.~~17. The recombinant polypeptide of claim 16 ~~43 or claim 44~~, comprising from N-terminus to C-terminus:

the protein of interest; the protease cleavage site; the protease; and the protein localization tag; or

the protein localization tag; the protease the protease cleavage site; and the protein of interest

.

~~46. The recombinant polypeptide of claim 43 or claim 44, comprising from N-terminus to C-terminus:~~

~~the protein localization tag;~~

~~the protease~~

~~the protease cleavage site; and~~

~~the protein of interest.~~

~~47.~~18. A nucleic acid or expression vector encoding the recombinant polypeptide of any one of claims 1 to 17~~46~~.

~~48. An expression vector comprising the nucleic acid of claim 47.~~

~~49.~~19. A cell comprising the nucleic acid ~~of claim 47~~ or the expression vector of claim ~~4~~18.

~~50.~~20. The cell of claim ~~4~~19, wherein the cell is a human T ~~mammalian~~ cell.

~~51. The cell of claim 50, wherein the cell is a human cell.~~

~~52. The cell of any one of claims 49 to 51, wherein the cell is an immune cell.~~

~~53. The cell of claim 52, wherein the immune cell is selected from the group consisting of: a T cell, a B cell, a natural killer (NK) cell, a macrophage, a monocyte, a neutrophil, a dendritic cell, a mast cell, a basophil, and an eosinophil.~~

~~54. The cell of claim 53, wherein the immune cell is a T cell.~~

~~55. The cell of claim 54, wherein the T cell is selected from the group consisting of: a naive T cell (TN), a cytotoxic T cell (Tcm), a memory T cell (THEM), a T memory stem cell (Tscm), a central memory T cell (Tcm), an effector memory T cell (TEM), a tissue resident memory T cell (TRH), an effector T cell (TEFF), a regulatory T cell (TREGS), a helper T cell, a CD4+ T cell, a CD8+ T cell, a virusspecific T cell, an alpha beta T cell (Tap), and a gamma delta T cell (Two).~~

~~56. The cell of claim 54 or claim 55, wherein the protein of interest is a CAR.~~

~~57. The cell of claim 56, wherein the protein of interest is the CAR of the recombinant polypeptide of any one of claims 15 to 22.~~

~~58. The cell of claim 54, wherein the protein of interest is a TCR.~~

~~59. The cell of any one of claims 49 to 58, wherein the nucleic acid encodes the recombinant polypeptide of any one of claims 43 to 46.~~

~~60.~~21. A ~~The~~ cell comprising a nucleic acid ~~of any one of claims 49 to 58,~~ encoding the recombinant polypeptide of any one of claims 1 to 17 or an expression vector encoding the recombinant polypeptide of any one of claims 1 to 17, wherein the recombinant polypeptide does not comprise a protease, and wherein the cell further comprises a nucleic acid encoding a protease, wherein the protease cleavage site is a cleavage site for the protease.

~~61. The cell of claim 60, further comprising a nucleic acid encoding a protease, wherein the protease cleavage site is a cleavage site for the protease.~~

~~62. The cell of claim 61, wherein the nucleic acid encoding the protease is present on the expression vector of claim 48.~~

~~63. The cell of claim 61, wherein the nucleic acid encoding the protease is present on an expression vector other than the expression vector of claim 48.~~

~~64.~~22. The cell of claim 21~~any one of claims 61 to 63~~, wherein the protease is (a) a soluble cytosolic protease or (b) is expressed on the cytosolic side of the cellular compartment determined by the protein localization tag.

~~65. The cell of any one of claims 61 to 63, wherein the protease is expressed on the cytosolic side of the cellular compartment determined by the protein localization tag.~~

~~66.~~23. The cell of claim 21 ~~any one of claims 61 to 63~~, wherein the protease is expressed on the lumen side of the cellular compartment determined by the protein localization tag.

~~67. A method of making the cell of any one of claims 49 to 66, comprising introducing the nucleic acid of claim 47 or the expression vector of claim 48 into the cell.~~

~~68. A method of making the recombinant polypeptide of any one of claims 1 to 46, comprising culturing a cell comprising the expression vector of claim 48 under conditions in which the cell expresses the recombinant polypeptide.~~

~~69.~~24. A method of regulating cellular localization of a protein of interest, comprising:

contacting ~~a~~ the cell of any one of claims 19 to 23 ~~that expresses:~~

~~the recombinant polypeptide of any one of claims 1 to 42; and~~

~~a protease, wherein the protease cleavage site is a cleavage site for the protease,~~

with an inhibitor of the protease when retention of the protein of interest at the cellular compartment determined by the protein localization tag is desired.

~~70. The method according to claim 69, wherein the cellular compartment determined by the protein localization tag is selected from the group consisting of: ER, Golgi, lysosome, plasma membrane, mitochondria, peroxisome, cytosol, and nucleus.~~

~~71. The method according to claim 69 or claim 70, wherein the protein of interest is engineered.~~

~~72. The method according to claim 71, wherein the engineered protein of interest is an engineered receptor.~~

~~73.~~25. The method according to claim 24~~72~~, wherein the method comprises regulating cellular localization of the ~~engineered receptor~~ protein of interest between the cellular compartment determined by the protein localization tag and the cell surface.

~~74. The method according to claim 72 or claim 73, wherein the cellular compartment determined by the protein localization tag is selected from the group consisting of: ER, Golgi, and lysosome.~~

~~75. The method according to any one of claims 72 to 74, wherein the engineered receptor is a CAR.~~

~~76. The method according to any one of claims 72 to 74, wherein the engineered receptor is a TCR.~~

~~77. The method according to any one of claims 69 to 71, wherein the protein of interest is a transcription factor.~~

~~78. The method according to claim 77, wherein the method comprises regulating cellular localization of the transcription factor between the cellular compartment determined by the protein localization tag and the nucleus.~~

~~79. The method according to claim 78, wherein the cellular compartment determined by the protein localization tag is the plasma membrane.~~

~~80. The method according to claim 78, wherein the cellular compartment determined by the protein localization tag is the cytosol.~~

~~81. The method according to claim 78, wherein the cellular compartment determined by the protein localization tag is selected from the group consisting of: ER, Golgi, and lysosome.~~

~~82. The method according to any one of claims 69 to 71, wherein the protein of interest is a secreted effector molecule.~~

~~83. The method according to claim 82, wherein the secreted effector molecule is selected from the group consisting of: a stimulatory ligand, an inhibitory ligand, a cytokine, a chemokine, a growth factor, and a protease.~~

~~84. The method according to claim 82 or claim 83, wherein the protein localization tag is an ER localization tag.~~

~~85. The method according to claim 84, wherein the secreted effector molecule is insoluble and positioned in the ER lumen in the presence of the protease inhibitor.~~

~~86. The method according to claim 85, wherein upon ceasing the contacting, the secreted effector molecule becomes soluble in the ER lumen and is secreted into the extracellular space.~~

~~87.~~26. The method of claim 24 or 25~~according to any one of claims 69 to 81~~, wherein the protease is derived from HCV NS3, and wherein the inhibitor of the protease is selected from the group consisting of: asunaprevir (ASV), danoprevir (DPV), simeprevir (SPV), grazoprevir (GPV), and any combination thereof.

~~88.~~27. The method according to any one of claims 24 to 26 ~~69 to 87~~, further comprising ceasing the contacting when retention of the protein of interest at the cellular compartment determined by the protein localization tag is no longer desired.

~~89. The method according to any one of claims 69 to 88, wherein the method is performed in vitro.~~

~~90. The method according to any one of claims 69 to 88, wherein the method is performed ex vivo.~~

~~91. The method according to any one of claims 69 to 88, wherein the method is performed in vivo.~~

~~92. The method according to any one of claims 69 to 91, wherein the recombinant polypeptide comprises the protease.~~

~~93. The method according to any one of claims 69 to 91, wherein the recombinant polypeptide does not comprise the protease.~~

~~94.~~28. A pharmaceutical composition comprising:

the cell of any one of claims 19 to 23 ~~49 to 66~~; and

a pharmaceutically acceptable carrier.

~~95. A method of making the pharmaceutical composition of claim 94, comprising introducing the expression vector of claim 48 into cells obtained from an individual.~~

~~96.~~29. A method of administering a regulatable cell-based therapy to an individual in need thereof, comprising administering to the individual the pharmaceutical composition of claim 28 ~~94, wherein the cells express a protease, and wherein the protease cleavage site is a cleavage site for the protease~~.

~~97. The method according to claim 96, wherein the recombinant polypeptide comprises the protease.~~

~~98. The method according to claim 96, wherein the recombinant polypeptide does not comprise the protease.~~

~~99.~~30. The method of claim 29 ~~according to any one of claims 96 to 98~~, further comprising administering to the individual an inhibitor of the protease when retention of the protein of interest at the cellular compartment determined by the protein localization tag is desired.

~~100.~~31. The method according to claim 30~~99~~, wherein the inhibitor of the protease is administered concurrently with the pharmaceutical composition or subsequently to administration of the pharmaceutical composition.

~~101. The method according to claim 99 or claim 100, wherein the inhibitor of the protease is administered subsequently to administration of the pharmaceutical composition.~~

~~102.~~32. The method according to any one of claims 29 to 31 ~~99 to 101~~, further comprising ceasing administration of the protease inhibitor when retention of the protein of interest at the cellular compartment determined by the protein localization tag is no longer desired.

~~103. The method according to any one of claims 99 to 102, wherein the protease is derived from HCV NS3, and wherein the protease inhibitor is selected from the group consisting of: asunaprevir (ASV), danoprevir (DPV), simeprevir (SPV), grazoprevir (GPV), and any combination thereof.~~

~~104. The method according to any one of claims 96 to 103, wherein the pharmaceutical composition comprises immune cells comprising the expression vector of claim 48.~~

~~105. The method according to claim 104, wherein the immune cells are selected from the group consisting of: T cells, B cells, natural killer (NK) cells, macrophages, monocytes, neutrophils, dendritic cells, mast cells, basophils, and eosinophils.~~

106. The method according to claim 105, wherein the immune cells are T cells.

107. The method according to claim 106, wherein the protein of interest is a CAR.

108. The method according to claim 106, wherein the protein of interest is a TCR.

109. A kit, comprising:
the expression vector of claim 48; and
instructions for introducing the expression vector into a cell.

110. The kit of claims 109, wherein the instructions further comprise instructions for regulating cellular localization of the protein of interest.

111. The kit of claim 110, wherein the instructions comprise instructions for contacting the cell or progeny thereof with an inhibitor of the protease when retention of the protein of interest at the cellular compartment determined by the protein localization tag is desired.

112. The kit of claim 111, wherein the instructions comprise instructions for ceasing the contacting when retention of the protein of interest at the cellular compartment determined by the protein localization tag is no longer desired.

113. The kit of any one of claims 109 to 112, further comprising an inhibitor of the protease.