CLAIMS

1. A method for expressing a plurality of target polypeptides at a high level, the method comprising:

(a) culturing a plurality of mammalian host cells, each comprising a plurality of recombinant polynucleotides, wherein

each recombinant polynucleotide comprises a promoter, a nucleotide sequence encoding a unique cell surface marker polypeptide, and a nucleotide sequence encoding a unique target polypeptide, wherein both the nucleotide sequence encoding the unique cell surface marker polypeptide and the nucleotide sequence encoding the unique target polypeptide are transcribed on the same mRNA,

wherein the culturing is under conditions that permit expression of each unique cell surface marker polypeptide on the surface of the mammalian host cells and expression of each unique target polypeptide;

(b) contacting the cultured mammalian host cells of step (a) with a plurality of detectable agents, each uniquely capable of binding one or more of the unique cell surface marker polypeptides expressed on the surface of the mammalian host cells;

(c) performing at least one round of fluorescence-activated cell sorting on the contacted cells from step (b), thereby selecting one or more mammalian host cells that are uniquely bound by at least one of the plurality of detectable agents;

(d) preparing one or more clonal populations of the mammalian host cells selected in step (c);

(e) analyzing one or more clonal populations from step (d) by detecting level of expression of at least two unique cell surface marker polypeptides on each of said clonal populations;

(f) selecting one or more clonal populations having high expression level of the at least two unique cell surface marker polypeptides; and

(g) culturing one or more clonal populations selected in step (f) under conditions that permit expression of the plurality of target polypeptides at a high level.

2.~~-~~ The method of claim 1, wherein step (c) comprises performing a single round of fluorescence-activated cell sorting on the contacted cells from step (b) with at least a first detectable agent and a second detectable agent, thereby selecting one or more mammalian host cells that are uniquely bound by at least both the first detectable agent and the second detectable agent~~.~~; or

~~3. The method of claim 1,~~ wherein step (c) comprises

~~(c~~ (c1) performing a first round of fluorescence-activated cell sorting on at least a first cell surface marker polypeptide, thereby selecting one or more mammalian host cells that are bound by at least a first detectable agent; and

(c2) performing a second round of fluorescence-activated cell sorting, on cells selected in step (~~c~~c1), on at least a second cell surface marker polypeptide, thereby selecting one or more mammalian host cells that are bound by at least both the first detectable agent and the second detectable agent.

~~4.~~

3. The method of ~~any one of claims~~claim 1 ~~to 3~~or 2, wherein:

(a) the step (e) is performed 7-28 days after step (d~~).~~);

~~5. The method of any one of claims 1 to 4, wherein~~(b) the analyzing in step (e) comprises flow cytometry~~.~~;

~~6. The method of any one of claims 1 to 5, wherein~~(c) the expression level of at least one of the plurality of unique cell surface marker polypeptides in step (f) is higher than a corresponding expression level of at least 70 % of the clonal populations analyzed in step (e~~).~~);

~~7. The method of any one of claims 1 to 5, wherein~~(d) the expression level of each of the plurality of unique cell surface marker polypeptides in step (f) is higher than a corresponding expression level of at least 70 % of the clonal populations analyzed in step (e~~).~~);

~~8. The method of any one of claims 1 to 5, wherein~~(e) the expression level of at least a first of the plurality of unique cell surface polypeptides in step (f) is higher than the expression level of at least a second of the plurality of unique cell surface polypeptides in step (f~~).~~); and/or

9. The method of claim 8,(f) wherein the expression level of a first of the plurality of unique cell surface polypeptides in step (f) is higher than the expression level of a second of the plurality of unique cell surface polypeptides in step (f).

10.

4. The method of any one of claims 1 to 93, further comprising:

(h) isolating at least a first unique target polypeptide and a second unique target polypeptide expressed in step (g) from the one or more selected clonal populations or from cell culture medium in which the one or more selected clonal populations are cultured.

11.

5. The method of any one of claims 1 to 104, wherein the plurality of unique target polypeptides comprises 2 to 8 unique target polypeptides., optionally

12. The method of claim 11, wherein the plurality of unique target polypeptides comprises 2 to 4 unique target polypeptides.,

13. The method of claim 12, wherein the plurality of unique target polypeptides comprises 2 unique target polypeptides.,

14. The method of claim 12, wherein the plurality of unique target polypeptides comprises 3 unique target polypeptides., or

15. The method of claim 12, wherein the plurality of unique target polypeptides comprises 4 unique target polypeptides.

16.

6. The method of any one of claims 1 to 155, wherein

each recombinant polynucleotide comprises, from 5' to 3', the promoter, the nucleotide sequence encoding the unique cell surface marker polypeptide, and the nucleotide sequence encoding the unique target polypeptide.;

17. The method of any one of claims 1 to 15, wherein

each recombinant polynucleotide comprises 1 to 4 promoters, 1 to 4 nucleotide sequences each encoding a unique cell surface marker polypeptide, and 1 to 4 nucleotide sequences each encoding a unique target polypeptide, wherein one of the nucleotide sequences encoding a unique cell surface marker polypeptide is transcribed on the same mRNA as one of the nucleotide sequences encoding a unique target polypeptide.;

~~18. The method of claim 17, wherein~~

each recombinant polynucleotide is a bicistronic polynucleotide comprising two promoters and two nucleotide sequences each encoding a unique target polypeptide, and further comprising one or two nucleotide sequences each encoding a unique cell surface marker polypeptide~~.~~;

~~19. The method of claim 18, wherein~~ each recombinant polynucleotide is a bicistronic polynucleotide comprising two promoters and two nucleotide sequences each encoding a unique target polypeptide, and further comprising one or two nucleotide sequences each encoding a unique cell surface marker polypeptide;

each bicistronic polynucleotide comprises a single nucleotide sequence encoding a unique cell surface marker polypeptide~~.~~;

~~20. The method of claim 18, wherein~~ each bicistronic polynucleotide comprises two nucleotide sequences each encoding a unique cell surface marker polypeptide, and wherein each one of the nucleotide sequences encoding a unique cell surface marker polypeptide is transcribed on the same mRNA as one of the nucleotide sequences encoding a unique target polypeptide~~.~~;

~~21. The method of any one of claims 17-20, wherein~~

each recombinant polynucleotide comprises, from 5' to 3', one of the from 1-4 promoters, one of the from 1-4 nucleotide sequences encoding a unique cell surface marker polypeptide, and one of the from 1--4 nucleotide sequences encoding a unique target polypeptide~~.~~;

~~22. The method of claim 17, wherein~~ each recombinant polynucleotide comprises one promoter, one nucleotide sequence encoding a unique cell surface marker polypeptide, and one nucleotide sequence encoding a unique target polypeptide, wherein the nucleotide sequence encoding a unique cell surface marker polypeptide is transcribed on the same mRNA as the nucleotide sequence encoding a unique target polypeptide~~.~~;

~~23. The method of claim 16 or 21, wherein~~ each recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3' to the nucleotide sequence encoding the unique cell surface marker polypeptide and 5' to the nucleotide sequence encoding the unique target polypeptide~~.~~; and/or

~~24. The method of any one of claims 1-23, wherein~~ each recombinant polynucleotide further comprises an alternate (non-~~AT G~~ATG) start codon for translation initiation of the nucleotide

sequence encoding the unique cell surface marker polypeptide and an ATG start codon for translation initiation of the nucleotide sequence encoding the unique target polypeptide.

25. The method of claim 24,, optionally wherein each alternate (non-ATG) start codon is selected independently from the group consisting of CTG, GTG, TTG, ATT, ATA, and ACG.,

26. The method of claim 24, wherein each alternate (non-ATG) start codon is selected independently from the group consisting of GTG and TTG.

27. The method of any one of claims 24 to 26,, or wherein the nucleotide sequence encoding the unique cell surface marker polypeptide is devoid of any ATG triplets.

28.

7. The method of any one of claims 1 to 156, wherein

at least a first recombinant polynucleotide comprises, from 5' to 3', a first promoter, a nucleotide sequence encoding a first unique cell surface marker polypeptide, and a nucleotide sequence encoding a first unique target polypeptide; and optionally wherein

at least a second recombinant polynucleotide comprises, from 5' to 3', a second promoter, a nucleotide sequence encoding a second unique target polypeptide, and a nucleotide sequence encoding a second unique cell surface marker polypeptide.

29. The method of claim 28, wherein the first recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3' to the nucleotide sequence encoding the first unique cell surface marker polypeptide and 5' to the nucleotide sequence encoding the first unique target polypeptide., or

30. The method of claim 28, wherein the first recombinant polynucleotide further comprises an alternate (non-ATG) start codon for translation initiation of the nucleotide sequence encoding the first unique cell surface marker polypeptide and an ATG start codon for translation initiation of the nucleotide sequence encoding the first unique target polypeptide.; and

31. The method of claim 30, wherein the alternate (non-ATG) start codon is selected at least a second recombinant polynucleotide comprises, from the group consisting of CTG, GTG, TTG, ATT, ATA, and ACG.

32. The method of claim 31, wherein the alternate (non-ATG) start codon is selected from the group consisting of GTG and TTG.

33. The method of any one of claims 285' to 32, wherein the3', a second promoter, a nucleotide sequence encoding the firsta second unique target polypeptide, and a nucleotide sequence encoding a second unique cell surface marker polypeptide is devoid of any ATG triplets., optionally wherein

~~34. The method of claim 28, wherein~~ the second recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3' to the nucleotide sequence encoding the second unique target polypeptide and 5' to the nucleotide sequence encoding the second unique cell surface marker polypeptide~~.~~; and/or

~~35. The method of any one of claims 1 to 15,~~ wherein

each recombinant polynucleotide comprises, from 5' to 3', the promoter, the nucleotide sequence encoding the unique target polypeptide, and the nucleotide sequence encoding the unique cell surface marker polypeptide~~.~~

~~36. The method of claim 35,~~, optionally wherein each recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3' to the nucleotide sequence encoding the unique target polypeptide and 5' to the nucleotide sequence encoding the unique cell surface marker polypeptide.

~~37.~~

8. The method of any one of claims 1 to ~~36~~7, wherein at least one promoter is a ~~0~~-β-actin promoter~~.~~

~~38. The method of any one of claims 1 to 36, wherein~~ , each promoter is a ~~0~~-β-actin promoter~~.~~

~~39. The method of any one of claims 1 to 36, wherein,~~ at least one promoter is a hamster ~~0~~-β-actin promoter~~.~~

~~40. The method of any one of claims 1 to 36, wherein~~, or each promoter is a hamster ~~0~~-β-actin promoter.

~~41.~~

9. The method of any one of claims 1 to ~~40~~8, wherein

at least a first unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, CD59, and variants thereof~~.~~;

~~42. The method of any one of claims 1 to 41, wherein~~ at least a second unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, CD59, and variants thereof~~.~~;

~~43. The method of any one of claims 1 to 42, wherein~~ each unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, CD59, and variants thereof~~.~~;

~~44. The method of any one of claims 1 to 40, wherein~~ at least a first unique cell surface marker polypeptide is selected from the group consisting of human CD52 and variants thereof~~.~~;

~~45. The method of any one of claims 1 to 40 and 44, wherein~~ at least a second unique cell surface marker polypeptide is selected from the group consisting of human CD52 and variants thereof~~.~~;

~~46. The method of claim 45, wherein~~ each unique cell surface marker polypeptide is selected from the group consisting of human CD52 and variants thereof~~.~~;

~~47. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, and CD59.~~

~~48. The method of any one of claims 1 to 40 and 47, wherein at least a second unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, and CD59.~~

~~49. The method of claim 48, wherein each unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, and CD59.~~

~~50. The method of any one of claims 1 to 40, wherein~~ at least a first unique cell surface marker polypeptide is human CD52~~.~~; and/or

~~51. The method of any one of claims 1 to 40, wherein~~ at least a second unique cell surface marker polypeptide is human CD52.

~~52.~~

10. The method of any one of claims 1 to ~~40~~9, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising:

(a) the amino acid sequence set forth in SEQ ID NO: 19 (GX_2NDTSQTSSPS) wherein X_2 is selected from the group consisting of A, G, I, L, and V~~.~~, optionally wherein X_2 is A;

~~53. The method of claim 52, wherein X2 is A.~~

~~54. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(b) the amino acid sequence set forth in SEQ ID NO: 20 (GQNX_4TSQTSSPS) wherein X_4 is selected from the group consisting of A, G, I, L, and V~~.~~, optionally wherein X_4 is A;

~~55. The method of claim 54, wherein X4 is A.~~

~~56. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(c) the amino acid sequence set forth in SEQ ID NO: 21 (GQNDTSX_7TSSPS) wherein X_7 is selected from the group consisting of A, G, I, L, and V~~.~~, optionally wherein X_7 is A;

~~57. The method of claim 56, wherein X7 is A.~~

~~58. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(d) the amino acid sequence set forth in SEQ ID NO: 22 (~~GQNDTSQXsSSPS~~GQNDTSQX_8SSPS) wherein ~~Xs~~X_8 is selected from the group consisting of A, G, I, L, and V~~.~~, optionally wherein X_8 is A;

~~59. The method of claim 58, wherein Xs is A.~~

~~60. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(e) the amino acid sequence set forth in SEQ ID NO: 23 (~~GQNDTSQXsXgSPS~~GQNDTSQX_8X_9SPS), wherein each of ~~Xs~~X_8 and X_9 is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~61. The method of claim 60,~~, optionally wherein each of ~~Xs~~X_8 and X_9 is A~~.~~;

~~62. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(f) the amino acid sequence set forth in SEQ ID NO: 24 (~~GQNDTSQXsSXioPS~~GQNDTSQX_8SX_{10}PS), wherein each of ~~Xs~~X_8 and X_{10} is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~63. The method of claim 62,~~, optionally wherein each of ~~Xs~~X_8 and X_{10} is A~~.~~

~~64. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(g) the amino acid sequence set forth in SEQ ID NO: 25 (~~GQNDTSQXSX9X10PS~~GQNDTSQ$X_8X_9X_{10}$PS), wherein each of ~~Xs~~X_8, X_9, and X_{10} is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~65. The method of claim 64,~~, optionally wherein each of ~~Xs~~X_8, X_9, and X_{10} is A~~.~~;

~~66. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(h) the amino acid sequence set forth in SEQ ID NO: 26 (~~GX2NDTSQXSX9SPS~~GX_2NDTSQX_8X_9SPS), wherein each of X_2, ~~Xs~~X_8, and X_9 is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~67. The method of claim 66,~~, optionally wherein each of X_2, ~~Xs~~X_8, and X_9 is A~~.~~;

~~68. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(i) the amino acid sequence set forth in SEQ ID NO: 27 (~~GX2NDTSQXSSX10PS~~GX_2NDTSQX_8SX_{10}PS), wherein each of X_2, ~~Xs~~X_8, and X_{10} is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~69. The method of claim 68,~~, optionally wherein each of X_2, ~~Xs~~X_8, and X_{10} is A~~.~~;

~~70. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(j) the amino acid sequence set forth in SEQ ID NO: 28 (GQNDTS$X_7X_8X_9$SPS), wherein each of X_7, ~~Xs~~X_8, and X_9 is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~71. The method of claim 70,~~, optionally wherein each of X_7, ~~Xs~~X_8, and X_9 is A~~.~~; or

~~72. The method of any one of claims 1 to 40, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising~~(k) the amino acid sequence set forth in SEQ ID NO: 29 (~~GQNDTSX7XSSX10PS~~GQNDTSX$_7$X$_8$SX$_{10}$PS), wherein each of X$_7$, ~~Xs~~X$_8$, and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V~~.~~

~~73. The method of claim 72,~~, optionally wherein each of X$_7$, ~~Xs~~X$_8$, and X$_{10}$ is A.

~~74.~~

11. The method of any one of claims 1 to ~~40~~10, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52

(a) comprising the amino acid sequence set forth in SEQ ID NO: 19 (GX$_2$NDTSQTSSPS) wherein X$_2$ is selected from the group consisting of A, G, I, L, and V; and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 20 (GQNX$_4$TSQTSSPS) and SEQ ID NO: 21 (GQNDTSX$_7$TSSPS) wherein each of X$_4$ and X$_7$ is independently selected from the group consisting of A, G, I, L, and V~~.~~;

~~75. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth in SEQ ID NO: 19 (GX2NDTSQTSSPS) wherein X2 is selected from the group consisting of A, G, I, L, and V; and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of an amino acid sequence selected from the group consisting of SEQ ID NO: 20 (GQNX4TSQTSSPS) and SEQ ID NO: 21 (GQNDTSX7TSSPS) wherein each of X4 and X7 is independently selected from the group consisting of A, G, I, L, and V.~~

~~76. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52~~(b) comprising the amino acid sequence set forth in SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 4 (GQNATSQTSSPS) and SEQ ID NO: 7 (GQNDTSATSSPS~~).~~);

~~77. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide~~(c) consists of a variant human CD52 consisting of the amino acid sequence set forth in SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of an amino acid sequence selected from the group consisting of SEQ ID NO: 4 (GQNATSQTSSPS) and SEQ ID NO: 7 (GQNDTSATSSPS~~).~~)

~~78. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide~~(d) comprises a variant human CD52 comprising the amino acid sequence set

forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS~~).~~.);

~~79. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide~~(e) consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS~~).~~.);

~~80. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52~~(f) comprising the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 7 (GQNDTSATSSPS~~).~~.);

~~81. The method~~(g) consists of ~~any one~~a variant human CD52 consisting of ~~claims 1 to 40, wherein~~the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a ~~first~~second unique cell surface marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: ~~2 (GANDTSQTSSPS); and at least a second unique cell surface~~
~~marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO:~~ 7 (GQNDTSATSSPS~~).~~.);

~~82. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide~~(h) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS); and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 7 (GQNDTSATSSPS~~).~~.);

~~83. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide~~(i) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 20 (GQNX_4TSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 23 (GX_2NDTSQX_8X_9SPS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 28 (GQNDTS$X_7X_8X_9$SPS), wherein each of X_4, X_7, ~~Xs~~X_8, and X_9 ~~_~~is independently selected from the group consisting of A, G, I, L, and V~~.~~;

84. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52(j) comprising the amino acid sequence set forth as SEQ ID NO: 20 (GQNX$_4$TSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 27 (GX$_2$NDTSQX$_8$SX$_{10}$PS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 29 (GQNDTSXyXsSXioPSGQNDTSX$_7$X$_8$SX$_{10}$PS) wherein each of X$_4$, X$_7$, X$_8$, and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V.;

85. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52(k) comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 15 (GANDTSQAASPS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 17 (GQNDTSAAASPS).); or

86. The method of any one of claims 1 to 40, wherein at least a first unique cell surface marker polypeptide(l) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 16 (GANDTSQASAPS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 18 (GQNDTSAASAPS).

12.87. The method of any one of claims 1 to 8611, wherein at least one unique target polypeptide comprises a therapeutic polypeptide or a therapeutic protein., optionally wherein

88. The method of any one of claims 1 to 86, wherein at least one unique target polypeptide is a polypeptide of a multi-chain protein.

89. The method of any one of claims 1 to 86, wherein each unique target polypeptide is a polypeptide of a multi-chain protein.,

90. The method of any one of claims 1 to 86, wherein at least one unique target polypeptide is a polypeptide of an antibody.,

91. The method of any one of claims 1 to 86, wherein each unique target polypeptide is a polypeptide of an antibody.

92. The method of any one of claims 1 to 86, wherein at least one unique target polypeptide is a polypeptide of a crossover dual-variable domain (CODV) Ig-like protein.

93. The method of claim 92, wherein each unique target polypeptide is a polypeptide of a crossover dual-variable domain (CODV) Ig-like protein.

94. The method of claim 92, wherein at least one unique target polypeptide is a polypeptide of a CODV triabody., or

95. The method of claim 92, wherein each unique target polypeptide is a polypeptide of a CODV triabody.

13.96. The method of any one of claims 1 to 9512, wherein the recombinant mammalian host cell is a CHO cell.

14.97. An engineered mammalian host cell comprising a plurality of recombinant polynucleotides, wherein each recombinant polynucleotide comprises a promoter, a nucleotide sequence encoding a unique cell surface marker polypeptide, and a nucleotide sequence encoding a unique target polypeptide, wherein both the nucleotide sequence encoding the unique cell surface marker polypeptide and the nucleotide sequence encoding the unique target polypeptide are transcribed on the same mRNA.

15.98. The engineered mammalian host cell of claim 9715, wherein the plurality of unique target polypeptides comprises 2 to 8 unique target polypeptides.

99.

16. The engineered mammalian host cell of claim 98, wherein the plurality of unique target polypeptides comprises 2 to 4 unique target polypeptides.

100. The engineered mammalian host cell of claim 99, wherein the plurality of unique target polypeptides comprises 2 unique target polypeptides.

101. The engineered mammalian host cell of claim 99, wherein the plurality of unique target polypeptides comprises 3 unique target polypeptides.

102. The engineered mammalian host cell of claim 99, wherein the plurality of unique target polypeptides comprises 4 unique target polypeptides.

~~103. The engineered mammalian host cell of any one of claims 97 to 102~~14 or 15, wherein each recombinant polynucleotide comprises, from 5' to 3', the promoter, the nucleotide sequence encoding the unique cell surface marker polypeptide, and the nucleotide sequence encoding the unique target polypeptide~~.~~;

~~104. The engineered mammalian host cell of any one of claims 97 to 102, wherein~~

each recombinant polynucleotide comprises 1 to 4 promoters, 1 to 4 nucleotide sequences each encoding a unique cell surface marker polypeptide, and 1 to 4 nucleotide sequences each encoding a unique target polypeptide, wherein one of the nucleotide sequences encoding a unique cell surface marker polypeptide is transcribed on the same mRNA as one of the nucleotide sequences encoding a unique target polypeptide~~.~~;

~~105. The engineered mammalian host cell of claim 104, wherein~~

each recombinant polynucleotide is a bicistronic polynucleotide comprising two promoters and two nucleotide sequences each encoding a unique target polypeptide, and further comprising one or two nucleotide sequences each encoding a unique cell surface marker polypeptide;

each recombinant polynucleotide is a bicistronic polynucleotide comprising two promoters and two nucleotide sequences each encoding a unique target polypeptide, and further comprising one or two nucleotide sequences each encoding a unique cell surface marker polypeptide~~.~~;

~~106. The engineered mammalian host cell of claim 105, wherein~~ each bicistronic polynucleotide comprises a single nucleotide sequence encoding a unique cell surface marker polypeptide~~.~~;

~~107. The engineered mammalian host cell of claim 105, wherein~~ each bicistronic polynucleotide comprises two nucleotide sequences each encoding a unique cell surface marker polypeptide, and wherein each one of the nucleotide sequences encoding a unique cell surface marker polypeptide is transcribed on the same mRNA as one of the nucleotide sequences encoding a unique target polypeptide~~.~~;

~~108. The engineered mammalian host cell of any one of claims 104-107, wherein~~ each recombinant polynucleotide comprises, from 5' to 3', one of the from 1-4 promoters, one of the from 1-4 nucleotide sequences encoding a unique cell surface marker polypeptide, and one of the from 1--4 nucleotide sequences encoding a unique target polypeptide~~.~~;

~~109. The engineered mammalian host cell of claim 104, wherein~~ each recombinant polynucleotide comprises one promoter, one nucleotide sequence encoding a unique cell surface marker polypeptide, and one nucleotide sequence encoding a unique target polypeptide, wherein the nucleotide sequence encoding a unique cell surface marker polypeptide is transcribed on the same mRNA as the nucleotide sequence encoding a unique target polypeptide~~.~~;

~~110. The engineered mammalian host cell of claim 103 or 108, wherein~~ each recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3' to the nucleotide sequence encoding the unique cell surface marker polypeptide and 5' to the nucleotide sequence encoding the unique target polypeptide~~.~~; and/or

~~111. The engineered mammalian host cell of any one of claims 97-110, wherein~~ each recombinant polynucleotide further comprises an alternate (non-ATG) start codon for translation initiation of the nucleotide sequence encoding the unique cell surface marker polypeptide and an ATG start codon for translation initiation of the nucleotide sequence encoding the unique target polypeptide~~.~~, optionally wherein each alternate (non-ATG) start codon is selected independently from the group consisting of CTG, GTG, TTG, ATT, ATA, and ACG,

~~112. The engineered mammalian host cell of claim 111, wherein~~ each alternate (non-ATG) start codon is selected independently from the group consisting of ~~CTG,~~ GTG~~,~~ and TTG, ~~ATT, ATA, and ACG.~~

~~113. The engineered mammalian host cell of claim 111, wherein each alternate (non-ATG) start codon is selected independently from the group consisting of GTG and TTG.~~

~~114. The engineered mammalian host cell of any one of claims 111 to 112,~~or wherein the nucleotide sequence encoding the unique cell surface marker polypeptide is devoid of any ATG triplets.

~~115.~~

17. The engineered mammalian host cell of any one of claims ~~97~~14 to ~~103~~16, wherein at least a first recombinant polynucleotide comprises, from 5' to 3', a first promoter, a nucleotide sequence encoding a first unique cell surface marker polypeptide, and a nucleotide sequence encoding a first unique target polypeptide; ~~and~~optionally wherein

~~at least a second recombinant polynucleotide comprises, from 5' to 3', a second promoter, a nucleotide sequence encoding a second unique target polypeptide, and a nucleotide sequence encoding a second unique cell surface marker polypeptide.~~

~~116. The engineered mammalian host cell of claim 115, wherein~~ the first recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3' to the

nucleotide sequence encoding the first unique cell surface marker polypeptide and 5’ to the nucleotide sequence encoding the first unique target polypeptide., or

117. The engineered mammalian host cell of claim 115, wherein the first recombinant polynucleotide further comprises an alternate (non-ATG) start codon for translation initiation of the nucleotide sequence encoding the first unique cell surface marker polypeptide and an ATG start codon for translation initiation of the nucleotide sequence encoding the first unique target polypeptide.; and

118. The engineered mammalian host cell of claim 117, wherein the alternate (non-ATG) start codon is selected at least a second recombinant polynucleotide comprises, from the group consisting of CTG, GTG, TTG, ATT, ATA, and ACG.

119. The engineered mammalian host cell of claim 118, wherein the alternate (non-ATG) start codon is selected from the group consisting of GTG and TTG.

120. The engineered mammalian host cell of any one of claims 1155’ to 119, wherein the3’, a second promoter, a nucleotide sequence encoding the firsta second unique target polypeptide, and a nucleotide sequence encoding a second unique cell surface marker polypeptide is devoid of any ATG triplets., optionally wherein

121. The engineered mammalian host cell of claim 115, wherein the second recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3’ to the nucleotide sequence encoding the second unique target polypeptide and 5’ to the nucleotide sequence encoding the second unique cell surface marker polypeptide.; and/or

122. The engineered mammalian host cell of any one of claims 97 to 102, wherein each recombinant polynucleotide comprises, from 5’ to 3’, the promoter, the nucleotide sequence encoding the unique target polypeptide, and the nucleotide sequence encoding the unique cell surface marker polypeptide.

123. The engineered mammalian host cell of claim 122,, optionally wherein each recombinant polynucleotide further comprises an internal ribosome entry site (IRES) located 3’ to the nucleotide sequence encoding the unique target polypeptide and 5’ to the nucleotide sequence encoding the unique cell surface marker polypeptide.

124.

18. The engineered mammalian host cell of any one of claims 9714 to 123, wherein at least one promoter is a 0-actin promoter.

125. The engineered mammalian host cell of any one of claims 97 to 123, wherein each promoter is a 0-actin promoter.

126. The engineered mammalian host cell of any one of claims 97 to 123, wherein at least one promoter is a hamster 0 -actin promoter.

127. The engineered mammalian host cell of any one of claims 97 to 123, wherein each promoter is a hamster 0 -actin promoter.

128. The engineered mammalian host cell of any one of claims 97 to 12717, wherein at least a first unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, CD59, and variants thereof.

129. The engineered mammalian host cell of any one of claims 97 to 128, wherein at least a second unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, CD59, and variants thereof.;

130. The engineered mammalian host cell of any one of claims 97 to 129, wherein each unique cell surface marker polypeptide is selected from the group consisting of CD20, CD52, CD59, and variants thereof.;

131. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide is selected from the group consisting of human CD52 and variants thereof.;

132. The engineered mammalian host cell of any one of claims 97 to 127 and 131, wherein at least a second unique cell surface marker polypeptide is selected from the group consisting of human CD52 and variants thereof.;

133. The engineered mammalian host cell of claim 132, wherein each unique cell surface marker polypeptide is selected from the group consisting of human CD52 and variants thereof.;

134. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide is selected from the group consisting of CD20,human CD52,; and CD59./or

135. at least a second unique cell surface marker polypeptide is human CD52.

19. The engineered mammalian host cell of any one of claims 97 to 127 and 124,14 to 18, wherein at least a first unique cell surface marker polypeptide is a variant of CD52 comprising:

(a) the amino acid sequence set forth in SEQ ID NO: 19 (GX_2NDTSQTSSPS) wherein at least a second unique cell surface marker polypeptideX_2 is selected from the group consisting of A, G, I, L, and V, optionally wherein X_2 is A;

(b) the amino acid sequence set forth in SEQ ID NO: 20 (GQNX$_4$TSQTSSPS) wherein X$_4$ is selected from the group consisting of A, G, I, L, and V, optionally wherein X$_4$ is A;

(c) the amino acid sequence set forth in SEQ ID NO: 21 (GQNDTSX$_7$TSSPS) wherein X$_7$ is selected from the group consisting of ~~CD20, CD52~~A, G, I, L, and ~~CD59.~~V, optionally wherein X$_7$ is A;

~~136. The engineered mammalian host cell of claim 135, wherein each unique cell surface marker polypeptide~~(d) the amino acid sequence set forth in SEQ ID NO: 22 (GQNDTSQX$_8$SSPS) wherein X$_8$ is selected from the group consisting of ~~CD20, CD52~~A, G, I, L, and ~~CD59.~~

~~137. The engineered mammalian host cell of any one of claims 97 to 127,~~V, optionally wherein ~~at least a first unique cell surface marker polypeptide~~X$_8$ is ~~human CD52.~~A;

~~138. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a second unique cell surface marker polypeptide is human CD52.~~

~~139. The engineered mammalian host cell of any one~~(e) the amino acid sequence set forth in SEQ ID NO: 23 (GQNDTSQX$_8$X$_9$SPS), wherein each of X$_8$ and X$_9$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_8$ and X$_9$ is A;

(f) the amino acid sequence set forth in SEQ ID NO: 24 (GQNDTSQX$_8$SX$_{10}$PS), wherein each of X$_8$ and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_8$ and X$_{10}$ is A

(g) the amino acid sequence set forth in SEQ ID NO: 25 (GQNDTSQX$_8$X$_9$X$_{10}$PS), wherein each of X$_8$, X$_9$, and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_8$, X$_9$, and X$_{10}$ is A;

(h) the amino acid sequence set forth in SEQ ID NO: 26 (GX$_2$NDTSQX$_8$X$_9$SPS), wherein each of X$_2$, X$_8$, and X$_9$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_2$, X$_8$, and X$_9$ is A;

(i) the amino acid sequence set forth in SEQ ID NO: 27 (GX$_2$NDTSQX$_8$SX$_{10}$PS), wherein each of X$_2$, X$_8$, and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_2$, X$_8$, and X$_{10}$ is A;

(j) the amino acid sequence set forth in SEQ ID NO: 28 (GQNDTSX$_7$X$_8$X$_9$SPS), wherein each of X$_7$, X$_8$, and X$_9$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_7$, X$_8$, and X$_9$ is A; or

(k) the amino acid sequence set forth in SEQ ID NO: 29 (GQNDTSX$_7$X$_8$SX$_{10}$PS), wherein each of X$_7$, X$_8$, and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V, optionally wherein each of X$_7$, X$_8$, and X$_{10}$ is A.

20. The engineered mammalian host cell of any one of claims ~~97~~14 to ~~127~~19, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising:

(a) the amino acid sequence set forth in SEQ ID NO: 19 (GX$_2$NDTSQTSSPS) wherein X$_2$ is selected from the group consisting of A, G, I, L, and V.

~~140. The engineered mammalian host cell of claim 139, wherein X2 is A.~~

~~141. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 20 (GQNX4TSQTSSPS) wherein X4 is selected from the group consisting of A, G, I, L, and V.~~

~~142. The engineered mammalian host cell of claim 141, wherein X4 is A.~~

~~143. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 21 (GQNDTSX7TSSPS) wherein X7 is selected from the group consisting of A, G, I, L, and V.~~

~~144. The engineered mammalian host cell of claim 143, wherein X7 is A.~~

~~145. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 22 (GQNDTSQXsSSPS) wherein Xs is selected from the group consisting of A, G, I, L, and V.~~

~~146. The engineered mammalian host cell of claim 145, wherein Xs is A.~~

~~147. The engineered mammalian host cell of any one of claims 77-102, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 23 (GQNDTSQXsXgSPS), wherein each of Xs and X9 is independently selected from the group consisting of A, G, I, L, and V.~~

~~148. The engineered mammalian host cell of claim 147, wherein each of Xs and X9 is A.~~

~~149. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 24 (GQNDTSQXsSXioPS), wherein each of Xs and X10 is independently selected from the group consisting of A, G, I, L, and V.~~

~~150. The engineered mammalian host cell of claim 149, wherein each of Xs and X10 is A.~~

~~151. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid~~

~~sequence set forth in SEQ ID NO: 25 (GQNDTSQXSX9X10PS), wherein each of Xs, X9, and X10 is independently selected from the group consisting of A, G, I, L, and V.~~

~~152. The engineered mammalian host cell of claim 151, wherein each of Xs, X9, and X10 is A.~~

~~153. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 26 (GX2NDTSQX8X9SPS), wherein each of X2, Xs, and X9 is independently selected from the group consisting of A, G, I, L, and V.~~

~~154. The engineered mammalian host cell of claim 153, wherein each of X2, Xs, and X9 is A.~~

~~155. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 27 (GX2NDTSQX8SX10PS), wherein each of X2, Xs, and X10 is independently selected from the group consisting of A, G, I, L, and V.~~

~~156. The engineered mammalian host cell of claim 155, wherein each of X2, Xs, and X10 is A.~~

~~157. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 28 (GQNDTSX7X8X9SPS), wherein each of X7, Xs, and X9 is independently selected from the group consisting of A, G, I, L, and V.~~

~~158. The engineered mammalian host cell of claim 157, wherein each of X7, Xs, and X9 is A.~~

~~159. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least one unique cell surface marker polypeptide is a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 29 (GQNDTSX7X8SX10PS), wherein each of X7, Xs, and X10 is independently selected from the group consisting of A, G, I, L, and V.~~

~~160. The engineered mammalian host cell of claim 159, wherein each of X7, Xs, and X10 is A.~~

~~161. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth in SEQ ID NO: 19 (GX2NDTSQTSSPS) wherein X2 is selected from the group consisting of A, G, I, L, and V; and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 20 (GQNX4TSQTSSPS) and SEQ ID NO: 21 (GQNDTSX7TSSPS) wherein each of X4 and X7 is independently selected from the group consisting of A, G, I, L, and V.~~

~~162. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth in SEQ ID NO: 19 (GX2NDTSQTSSPS) wherein X2 is selected from the group consisting of A, G, I, L, and V; and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of an amino acid sequence selected from the group consisting of SEQ ID NO: 20 (GQNX4TSQTSSPS) and SEQ ID NO: 21 (GQNDTSX7TSSPS) wherein each of X4 and X7 is independently selected from the group consisting of A, G, I, L, and V.~~

~~163. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52~~(b) comprising the amino acid sequence set forth in SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second

unique cell surface marker polypeptide comprises a variant human CD52 comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 4 (GQNATSQTSSPS) and SEQ ID NO: 7 (GQNDTSATSSPS~~).~~.);

~~164. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(c) consists of a variant human CD52 consisting of the amino acid sequence set forth in SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of an amino acid sequence selected from the group consisting of SEQ ID NO: 4 (GQNATSQTSSPS) and SEQ ID NO: 7 (GQNDTSATSSPS~~).~~.)

~~165. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(d) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS~~).~~.);

~~166. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(e) consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS~~).~~.);

~~167. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52~~(f) comprising the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 7 (GQNDTSATSSPS~~).~~.);

~~168. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(g) consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: 2 (GANDTSQTSSPS); and at least a second unique cell surface marker polypeptide consists of a variant human CD52 consisting of the amino acid sequence set forth as SEQ ID NO: 7 (GQNDTSATSSPS~~).~~.);

~~169. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(h) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS); and at least a second

unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 7 (GQNDTSATSSPS~~).~~);

~~170. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(i) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 20 (GQNX$_4$TSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 23 (GX$_2$NDTSQX$_8$X$_9$SPS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 28 (GQNDTSX$_7$X$_8$X$_9$SPS), wherein each of X$_4$, X$_7$, ~~Xs~~X$_8$, and X$_9$ is independently selected from the group consisting of A, G, I, L, and V~~.~~;

~~171. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52~~(j) comprising the amino acid sequence set forth as SEQ ID NO: 20 (GQNX$_4$TSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 27 (GX$_2$NDTSQX$_8$SX$_{10}$PS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 29 (~~GQNDTSXyXsSXioPS~~GQNDTSX$_7$X$_8$SX$_{10}$PS) wherein each of X$_4$, X$_7$, ~~Xs~~X$_8$, and X$_{10}$ is independently selected from the group consisting of A, G, I, L, and V~~.~~;

~~172. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide comprises a variant human CD52~~(k) comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 15 (GANDTSQAASPS); and at least a third unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 17 (GQNDTSAAASPS~~).~~); or

~~173. The engineered mammalian host cell of any one of claims 97 to 127, wherein at least a first unique cell surface marker polypeptide~~(l) comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 4 (GQNATSQTSSPS); at least a second unique cell surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 16 (GANDTSQASAPS); and at least a third unique cell

surface marker polypeptide comprises a variant human CD52 comprising the amino acid sequence set forth as SEQ ID NO: 18 (GQNDTSAASAPS).

~~174.~~

21. The engineered mammalian host cell of any one of claims ~~97~~14 to ~~173~~21, wherein at least one unique target polypeptide comprises a therapeutic polypeptide or a therapeutic protein~~. 175. The engineered mammalian host cell of any one of claims 97 to 173, wherein at least one unique target polypeptide is a polypeptide of a multi-chain protein.~~, optionally wherein

~~176. The engineered mammalian host cell of any one of claims 97 to 173~~at least one unique target polypeptide is a polypeptide of a multi-chain protein, wherein each unique target polypeptide is a polypeptide of a multi-chain protein~~.~~,

~~177. The engineered mammalian host cell of any one of claims 97 to 173, wherein~~ at least one unique target polypeptide is a polypeptide of an antibody~~.~~,

~~178. The engineered mammalian host cell of any one of claims 97 to 173, wherein~~ each unique target polypeptide is a polypeptide of an antibody~~.~~,

~~179. The engineered mammalian host cell of any one of claims 97 to 173, wherein~~ at least one unique target polypeptide is a polypeptide of a crossover dual-variable domain (CODV) Ig-like protein~~.~~,

~~180. The engineered mammalian host cell of claim 179, wherein~~ each unique target polypeptide is a polypeptide of a crossover dual-variable domain (CODV) Ig-like protein~~.~~,

~~181. The engineered mammalian host cell of any one of claims 97 to 173, wherein~~ at least one unique target polypeptide is a polypeptide of a CODV triabody~~.~~, or

~~182. The engineered mammalian host cell of any one of claims 97 to 173, wherein~~ each unique target polypeptide is a polypeptide of a CODV triabody.

~~183. The engineered mammalian host cell of any one of claims 97 to 182, wherein the recombinant mammalian host cell is a CHO cell.~~

~~184.~~

22. An isolated variant human CD52 comprising an amino acid sequence selected from the group consisting of SEQ ID NO: 19 ($GX_2NDTSQTSSPS$) wherein X_2 is selected from the group consisting of A, G, I, L, and V~~.~~;

~~185. The isolated variant human CD52 of claim 184, wherein X2 is A.~~

~~186. An isolated variant human CD52 comprising an amino acid sequence consisting of~~

_____SEQ ID NO: 20 (GQNX$_4$TSQTSSPS) wherein X$_4$ is selected from the group consisting of A, G, I, L, and V.; or

~~187. The isolated variant human CD52 of claim 186, wherein X4 is A.~~
~~188. An isolated variant human CD52 comprising an amino acid sequence consisting of~~ ____SEQ ID NO: 22 (~~GQNDTSQXsSSPS~~GQNDTSQX$_8$SSPS) wherein ~~Xs~~X$_8$ is selected from the group consisting of A, G, I, L, and V.