

Claims:

1. A specific binding molecule having the property of binding to KVLEYVIKV (SEQ ID NO: 1) HLA-A*02 complex and/or KVLEYVIKV (SEQ ID No: 17) HLA-A*02 complex and comprising a TCR alpha chain variable domain and/or a TCR beta chain variable domain each of which comprises FR1-CDR1-FR2-CDR2-FR3-CDR3-FR4 where FR is a framework region and CDR is a complementarity determining region, wherein
- the alpha chain CDRs have the following sequences:
- CDR1 – SSVPPY (SEQ ID NO: 15)
- CDR2 – YTSAATLV (SEQ ID NO: 16)
- CDR3 – AARPSSSNTGKLI (SEQ ID NO: 17)
- optionally with one or more mutations therein,
- and/or
- (b) the beta chain CDRs have the following sequences:
- CDR1 – PRHDT (SEQ ID NO: 18)
- CDR2 – FYEKMV (SEQ ID NO: 19)
- CDR3 – ASSFTGFDEQF (SEQ ID NO: 20)
- optionally with one or more mutations therein
2. The specific binding molecule of claim 1, wherein the alpha chain variable domain framework regions comprise the following sequences:
- FR1 – amino acids 1-26 of SEQ ID NO: 2
- FR2 – amino acids 33-49 of SEQ ID NO: 2
- FR3 – amino acids 58-91 of SEQ ID NO: 2
- FR4 – amino acids 105-115 of SEQ ID NO: 2
- or respective sequences having at least 90, at least 91, at least 92, at least 93, at least 94, at least 95, at least 96, at least 97, at least 98 or at least 99% identity to said sequences, and/or the beta chain variable domain framework regions may comprise the following sequences:
- FR1 – amino acids 1-26 of SEQ ID NO: 3
- FR2 – amino acids 32-48 of SEQ ID NO: 3
- FR3 – amino acids 55-91 of SEQ ID NO: 3
- FR4 – amino acids 103-112 of SEQ ID NO: 3
- or respective sequences having at least 90, at least 91, at least 92, at least 93, at least 94, at least 95, at least 96, at least 97, at least 98 or at least 99% identity to said sequences.
3. The specific binding molecule of claim 1 or claim 2, wherein the one or more of the mutations in the alpha chain CDRs are selected from (with reference to the numbering of SEQ ID NO: 2):
- Insertion of 4 amino acids (ARWG) after position 26
 - S27D
 - S28G

- S52G
- A53G
- A54D
- T55L
- 5 • I56V
- S97D
- S98A

4. The specific binding molecule of claim 3, wherein
 10 the alpha chain CDR1 comprises the following sequence: SSVPPY (SEQ ID NO: 15) or
 ARWGDGVPPY (SEQ ID NO: 21);
 the alpha chain CDR2 comprises the following sequence: Y TSAATLV (SEQ ID NO: 16) or
 YTGGDLVV (SEQ ID NO: 22); and/or
 the alpha chain CDR3 comprises the following sequence: AARPSSSNTGKLI (SEQ ID NO:
 15 17), AARPSDSNTGKLI (SEQ ID NO: 23) or AARPSSANTGKLI (SEQ ID NO: 24).

5. The specific binding molecule of any preceding claim, wherein the one or more of the
 mutations in the beta chain CDRs are selected from (with reference to the numbering of SEQ ID NO:
 3):

- 20 • Y50F
- K52T
- M53K
- Q54F
- F95V
- 25 • T96W
- G97D
- F98W/Y

6. The specific binding molecule of claim 5, wherein
 30 the beta chain CDR1 comprises the following sequence: PRHDT (SEQ ID NO: 18);
 the beta chain CDR2 comprises the following sequence: FYEKM (SEQ ID NO: 19),
 FFETMF (SEQ ID NO: 25) or FFETKF (SEQ ID NO: 26); and/or
 the beta chain CDR3 comprises the following sequence: ASSFTGFDEQF (SEQ ID NO: 20),
 ASSVWDWDEQF (SEQ ID NO: 27) or ASSVWDYDEQF (SEQ ID NO: 28).

35 7. The specific binding molecule of any preceding claim, which has one of the following
 combinations of alpha and beta CDRs:

	Alpha			Beta		
	CDR1	CDR2	CDR3	CDR1	CDR2	CDR3
a	<u>ARWGDGVPPY</u>	<u>YTGGDLVV</u>	<u>AARPSSSNTGKLI</u>	<u>PRHDT</u>	<u>FFETMF</u>	<u>ASSVWDWDEQF</u>
b	<u>ARWGDGVPPY</u>	<u>YTGGDLVV</u>	<u>AARPSSSNTGKLI</u>	<u>PRHDT</u>	<u>FFETKF</u>	<u>ASSVWDYDEQF</u>
c	<u>ARWGDGVPPY</u>	<u>YTGGDLVV</u>	<u>AARPSDSNTGKLI</u>	<u>PRHDT</u>	<u>FFETMF</u>	<u>ASSVWDWDEQF</u>
d	<u>ARWGDGVPPY</u>	<u>YTGGDLVV</u>	<u>AARPSDSNTGKLI</u>	<u>PRHDT</u>	<u>FFETMF</u>	<u>ASSVWDYDEQF</u>
e	<u>ARWGDGVPPY</u>	<u>YTGGDLVV</u>	<u>AARPSSANTGKLI</u>	<u>PRHDT</u>	<u>FFETMF</u>	<u>ASSVWDYDEQF</u>

8. The specific binding molecule of any preceding claim, wherein the alpha chain variable domain comprises any one of the amino acid sequences of SEQ ID NOs: 4-6 or a sequence having at least 90% identity thereto, and the beta chain variable domain comprises any one of the amino acid sequences of SEQ ID NOs: 7-9 or a sequence having at least 90% identity thereto.

9. The specific binding molecule of any preceding claim, which comprises one of the following alpha and beta chain variable domain pairs:

Alpha chain variable domain	Beta chain variable domain
SEQ ID No: 4	SEQ ID No: 7
SEQ ID No: 4	SEQ ID No: 8
SEQ ID No: 5	SEQ ID No: 7
SEQ ID No: 5	SEQ ID No: 9
SEQ ID No: 6	SEQ ID No: 9

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10. The specific binding molecule of any preceding claim, which is an alpha-beta heterodimer, having an alpha chain TRAC constant domain sequence and a beta chain TRBC1 or TRBC2 constant domain sequence.

11. The specific binding molecule of claim 10, wherein the alpha and beta chain constant domain sequences are modified by truncation or substitution to delete a native disulphide bond between Cys4 of exon 2 of TRAC and Cys2 of exon 2 of TRBC1 or TRBC2.

12. The specific binding molecule of claim 10 or claim 11, wherein the alpha and/or beta chain constant domain sequence(s) are modified by substitution of cysteine residues for Thr 48 of TRAC and Ser 57 of TRBC1 or TRBC2, the said cysteines forming a non-native disulphide bond between the alpha and beta constant domains of the TCR.

13. The specific binding molecule of any preceding claim, which is in single chain format of the type $V\alpha$ -L- $V\beta$, $V\beta$ -L- $V\alpha$, $V\alpha$ -C α -L- $V\beta$, $V\alpha$ -L- $V\beta$ -C β , wherein $V\alpha$ and $V\beta$ are TCR α and β variable

regions respectively, C α and C β are TCR α and β constant regions respectively, and L is a linker sequence.

14. The specific binding molecule of claim 13, wherein the linker sequence is selected from the group consisting of GGGGS (SEQ ID No: 29), GGGSG (SEQ ID No: 30), GGSGG (SEQ ID No: 31), GSGGG (SEQ ID No: 32), GSGGGP (SEQ ID No: 33), GGEPS (SEQ ID No: 34), GGEGGGP (SEQ ID No: 35), GGEGGGSEGGGS (SEQ ID No: 36), GGGSGGGG (SEQ ID NO: 37), GGGs (SEQ ID No: 38), GGGGS (SEQ ID No: 39), TVLRT (SEQ ID No: 40), TVSSAS (SEQ ID No: 41) and TVLSSAS (SEQ ID No: 42).
15. The specific binding molecule of any preceding claim associated with a detectable label, a therapeutic agent and/or a PK modifying moiety.
16. The specific binding molecule of claim 15, wherein an anti-CD3 antibody is covalently linked to the C- or N-terminus of the alpha or beta chain of the specific binding molecule, optionally via a linker sequence.
17. A specific binding molecule-anti-CD3 fusion molecule, comprising an alpha chain variable domain which comprises
 - an amino acid sequence selected from SEQ ID NOs: 4-6 or a sequence having at least 90% identity thereto,
 - a beta chain variable domain which comprises an amino acid sequence selected from SEQ ID NO: 7-9 or a sequence having at least 90% identity thereto, and
 - an anti-CD3 antibody, optionally comprising an amino acid sequence selected from SEQ NOS: 12-14, covalently linked to the N-terminus or C-terminus of the beta chain, optionally via a linker sequence.
18. The fusion molecule of claim 17, comprising:
 - an alpha variable domain having the sequence of SEQ ID NO 4 or a sequence at least 90% identical thereto and an alpha chain constant domain having the sequence of SEQ ID No 10 or a sequence at least 90% identical thereto, a beta variable domain having the sequence of SEQ ID NO 8 or a sequence at least 90% identical thereto and a beta chain constant domain having the sequence of SEQ ID No 11 or a sequence at least 90% identical thereto; and an anti-CD3 scFv antibody fragment having the sequence of SEQ ID NO 12 or a sequence at least 90% identical thereto fused to the N terminus of the beta chain via a linker having the sequence of SEQ ID NO: 29 or a sequence at least 90% identical thereto, or
 - an alpha variable domain having the sequence of SEQ ID NO 4 or a sequence at least 90% identical thereto and an alpha chain constant domain having the sequence of SEQ ID No 10 or a sequence at least 90% identical thereto, a beta variable domain having the sequence of SEQ ID NO 8 or a sequence at least 90% identical thereto, and a beta chain constant domain having the sequence of

SEQ ID No 11 or a sequence at least 90% identical thereto; and an anti-CD3 scFv antibody fragment having the sequence of SEQ ID NO 14 or a sequence at least 90% identical thereto fused to the N terminus of the beta chain via a linker having the sequence of SEQ ID NO: 29 or a sequence at least 90% identical thereto.

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19. A nucleic acid encoding an alpha chain and/or a beta chain as claimed in any one of the preceding claims.

20. An expression vector comprising the nucleic acid of claim 19.

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21. A cell harbouring

(a) an expression vector as claimed in claim 20 encoding alpha and beta variable chains as claimed in any one of claims 1 to 18, in a single open reading frame, or two distinct open reading frames; or

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(b) a first expression vector which comprises nucleic acid encoding the alpha variable chain of a specific binding molecule as claimed in any one of claims 1 to 18, and a second expression vector which comprises nucleic acid encoding the beta variable chain of a specific binding molecule as claimed in any one of claims 1 to 18.

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22. A non-naturally occurring and/or purified and/or engineered cell, especially a T-cell, presenting a specific binding molecule as claimed in any one of claims 1 to 16.

23. A pharmaceutical composition comprising a specific binding molecule as claimed in any one of claims 1 to 16, a specific binding molecule-anti CD3 fusion molecule as claimed in claim 17 or claim 18, a nucleic acid as claimed in claim 19 and/or a cell as claimed in claim 21 or claim 22, together with one or more pharmaceutically acceptable carriers or excipients.

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24. The specific binding molecule of any one of claims 1 to 16, specific binding molecule-anti-CD3 fusion molecule of claim 17 or claim 18, nucleic acid of claim 19, cell of claim 21 or claim 22, and/or pharmaceutical composition of claim 23 for use in medicine, preferably in a human subject.

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25. The specific binding molecule of any one of claims 1 to 16, specific binding molecule-anti-CD3 fusion molecule of claim 17 or claim 18, nucleic acid of claim 19, cell of claim 21 or claim 22, and/or pharmaceutical composition of claim 23 for use in for use in a method of treating cancer or a tumour, preferably in a human subject.

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26. A method of treating a human subject having cancer or a tumour, comprising administering to said subject in need thereof a pharmaceutically effective dose of a pharmaceutical composition according to claim 23.

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27. A method of producing a specific binding molecule according to any one of claims 1 to 16, or a specific binding molecule-anti-CD3 fusion molecule according to claim 17 or claim 18, comprising a) maintaining a cell according to claim 21 or 22 under optimal conditions for expression of the specific binding molecule chains and b) isolating the specific binding molecule chains.

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