CLAIMS

1. A polypeptide comprising, from N- to C-terminal:

a) a first single domain antigen binding domain (sdABD) that binds to a human tumor target antigen (TTA) (sdABD-TTA) and comprises the amino acid sequence of SEQ ID NOs:1, 5, 9, or 13;

b) a first domain linker;

c) a constrained Fv domain comprising:

i) a first variable heavy domain comprising a VHCDR1, VHCDR2 and VHCDR3;

ii) a constrained non-cleavable linker (CNCL); and

iii) a first variable light domain comprising VLCDR1, VLCDR2 and VLCDR3, wherein the CNCL is positioned between the first variable heavy domain and the first variable light domain and is too short to allow the first variable heavy domain and first variable light domain to bind each other, thereby preventing the first variable heavy domain and the first variable light domain from interacting to form an active Fv capable of binding CD3;

d) a second domain linker;

e) a second sdABD-TTA comprising the amino acid sequence of SEQ ID NOs:1, 5, 9, or 13;

f) a cleavable linker (CL);

g) a pseudo Fv domain comprising:

i) a first pseudo variable light domain, comprising an iVLCDR1, an iVLCDR2, and an iVLCDR3 that are inert, wherein the CDRs do not form antigen binding domains when self-associated;

ii) a non-cleavable linker (NCL); and

iii) a first pseudo variable heavy domain, comprising an iVHCDR1, an iVHCDR2 and an iVHCDR3 that are inert, wherein the CDRs do not form antigen binding domains when self-associated;

h) a third domain linker; and

i) a third sdABD that binds to human serum albumin (HSA);

wherein the first variable heavy domain and the first variable light domain are capable of binding human CD3, but the constrained Fv domain does not bind CD3; and the polypeptide does not bind CD3 when the CL is intact; and

wherein the first variable heavy domain and the first pseudo variable light domain intramolecularly associate to form an Fv that cannot bind CD3; and wherein the first variable light domain and the first pseudo variable heavy domain intramolecularly associate to form an Fv that cannot bind CD3.

2. A polypeptide comprising, from N- to C-terminal:

a) a first sdABD-TTA that comprises the amino acid sequence of SEQ ID NOs:1, 5, 9, or 13;

b) a first domain linker;

c) a constrained Fv domain comprising:

i) a first variable heavy domain comprising a VHCDR1, VHCDR2 and VHCDR3;

ii) a constrained cleavable linker (CCL); and

iii) a first variable light domain comprising VLCDR1, VLCDR2 and VLCDR3, wherein the CCL is positioned between the first variable heavy domain and the first variable light domain and is too short to allow the first variable heavy domain and first variable light domain to bind each other, thereby preventing the first variable heavy domain and the first variable light domain from interacting to form an active Fv capable of binding CD3;

d) a second domain linker;

e) a second sdABD-TTA that comprises the amino acid sequence of SEQ ID NOs:1, 5, 9, or 13;

f) a cleavable linker (CL);

g) a pseudo Fv domain comprising:

i) a first pseudo variable heavy domain, comprising an iVLCDR1, an iVLCDR2, and an iVLCDR3 that are inert, wherein the CDRs do not form antigen binding domains when self-associated;

ii) a non-cleavable linker (NCL); and

iii) a first pseudo variable light domain, comprising an iVHCDR1, an iVHCDR2 and an iVHCDR3 that are inert, wherein the CDRs do not form antigen binding domains when self-associated;

h) a third domain linker; and

i) a third sdABD that binds to human serum albumin;

wherein the first variable heavy domain and the first variable light domain are capable of binding human CD3, but the constrained Fv domain does not bind CD3; and the polypeptide does not bind CD3 when the CL is intact; and

wherein the first variable heavy domain and the first pseudo variable light domain intramolecularly associate to form an Fv that cannot bind CD3; and wherein the first variable light domain and the first pseudo variable heavy domain intramolecularly associate to form an Fv that cannot bind CD3.

3. A polypeptide according to any of claims 1 to 2 wherein the first variable heavy domain is N-terminal to the first variable light domain and the pseudo variable light domain is N-terminal to the pseudo variable heavy domain.

4. A polypeptide according to any of claims 1 to 2 wherein the first variable heavy domain is N-terminal to the first variable light domain and the pseudo variable heavy domain is N-terminal to the pseudo variable light domain.

5. A polypeptide according to any of claims 1 to 2 wherein the first variable light domain is N-terminal to the first variable heavy domain and the pseudo light variable light domain is N-terminal to the pseudo variable heavy domain.

6. A polypeptide according to any of claims 1 to 2 wherein the first variable light domain is N-terminal to the first variable heavy domain and the pseudo variable heavy domain is N-terminal to the pseudo variable light domain.

7. A polypeptide according to any of claims 1-6 wherein the first and second TTA is the same.

8. A polypeptide according to any of claims 1-6 wherein the first and second TTA are different.

9. A protein accordingly to any one of claims 1-8, wherein:

the vhCDR1 of the first variable heavy domain comprises the amino acid sequence of SEQ ID NO: 62;

the vhCDR2 of the first variable heavy domain comprises the amino acid sequence of SEQ ID NO: 63;

the vhCDR3 of the first variable heavy domain comprises the amino acid sequence of SEQ ID NO: 64;

the vlCDR1 of the first variable light domain comprises the amino acid sequence of SEQ ID NO: 50;

the vlCDR2 of the first variable light domain comprises the amino acid sequence of SEQ ID NO: 51; and

the vlCDR3 of the first variable light domain comprises the amino acid sequence of SEQ ID NO: 52.

10. A protein accordingly to any one of claims 1-9, wherein the first variable heavy domain comprises the amino acid sequence of SEQ ID NO: 61; and the first variable light domain comprises the amino acid sequence of SEQ ID NO: 49.

11. A protein accordingly to any one of claims 1-10, wherein:

the ivhCDR1 of the first pseudo heavy variable domain comprises the amino acid sequence of SEQ ID NO: 66 or 70;

the ivhCDR2 of the first pseudo heavy variable domain comprises the amino acid sequence of SEQ ID NO: 67 or 71;

the ivhCDR3 of the first pseudo heavy variable domain comprises the amino acid sequence of SEQ ID NO: 68 or 72;

the ivlCDR1 of the first pseudo light variable domain comprises the amino acid sequence of SEQ ID NO: 54 or 58;

the ivlCDR2 of the first pseudo light variable domain comprises the amino acid sequence of SEQ ID NO: 55 or 59; and

the ivlCDR3 of the first pseudo light variable domain comprises the amino acid sequence of SEQ ID NO: 56 or 60.

12. A protein according to any of claims 1-11, wherein the pseudo light variable domain comprises the amino acid sequence of SEQ ID NO:53; and the pseudo heavy variable domain comprises the amino acid sequence of SEQ ID NO:65.

13. A protein according to any of claims 1-11, wherein the pseudo light variable domain comprises the amino acid sequence of SEQ ID NO:57; and the pseudo heavy variable domain comprises the amino acid sequence of SEQ ID NO:69.

14. A polypeptide according to any of one of claims 1 to 13 wherein the third sdABD has the amino acid sequence of SEQ ID NO:45 or SEQ ID NO: 142.

15. A polypeptide according to claim 1 to 14 wherein the cleavable linker is cleaved by a human protease selected from the group consisting of MMP2, MMP9, Meprin A, Meprin B, Cathepsin S, Cathepsin K, Cathespin L, GranzymeB, uPA, Kallekriein7, matriptase and thrombin.

16. A polypeptide according to claim 1 comprising an amino acid sequence selected from the group consisting of SEQ ID NOs: 145, , 149, , 153, 169, 170, 154, 146, 165, 166, 167, 168, 157, 158, 159, 160, 162, and 163.

17. A protein according to any one of claims 1-16, comprising the amino acid sequence of SEQ ID NO: 145.

18. A protein according to any one of claims 1-16, comprising the amino acid sequence of SEQ ID NO: 149.

19. A nucleic acid encoding a polypeptide according to any of claims 1 to 18.

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

21. A host cell comprising the expression vector of claim 20.

22. A method of making a polypeptide comprising culturing the host cell of claim 21 under conditions wherein the polypeptide is expressed and recovering the polypeptide.

23. Use of a polypeptide of any of claims 1-18 in the manufacture of a medicament for the treatment of cancer.