

CLAIMS

1. A pair of polypeptide constructs comprising:

a) a first polypeptide comprising a first target antigen binding domain linked using a first domain linker to a first single chain Fv (ScFv) polypeptide directed to a CD-3 antigen, wherein said first polypeptide further comprises a half-life extension domain, and

b) a second polypeptide comprising a second target antigen binding domain linked using a second domain linker to a second scFv polypeptide directed to a CD-3 antigen, wherein said second polypeptide further comprises a half-life extension domain;

wherein said first scFv domain comprises a first VH domain and a first VL domain joined through a first scFv linker moiety comprising a first protease cleavage site between said first VH and said first VL domain, said first VH domain and said first VL domain interacting to form a first VH/VL pair, wherein said first VH domain is inactive (iVH) and said first VL domain is active (aVL), such that said first scFv domain does not specifically bind said CD-3 antigen;

wherein said second scFv domain comprises a second VH domain and a second VL domain joined through a second scFv linker moiety comprising a second protease cleavage site between said second VH and said second VL domain, said second VH domain and said second VL domain interacting to form a second VH/VL pair, wherein said second VL domain is inactive (iVL) and said second VH is active (aVH), such that said second scFv domain does not specifically bind said CD-3 antigen.

2. A pair of polypeptide constructs according to claim 1:

a) wherein said first polypeptide comprises, from N- to C-terminal:

i) said first target antigen binding domain;

ii) said first domain linker;

iii) said first scFv domain comprising:

1) said first active VL domain;

2) a cleavable linker; and

3) said first inactive VH domain;

v) a third domain linker; and

vi) said half-life extension domain; and

b) wherein said second polypeptide comprises, from N- to C-terminal:

- i) said second target antigen binding domain;
- ii) said second domain linker;
- iii) said second scFv domain comprising:
 - 1) said first active VH domain;
 - 2) a cleavable linker; and
 - 3) said first inactive VL domain;
- iv) a fourth domain linker; and
- v) said half-life extension domain.

3. A pair of polypeptides according to claim 1 or 2, wherein said half-life extension domain binds to human HSA.

4. A pair of polypeptides according to any previous claim wherein said half-life extension domain has the sequence

EVQLVESGGGLVQPGNSLRRLSCAASGFTFSKFGMSWVRQAPGKGLEWVSSISGSGRDTLYAESV
KGRFTISRDNAAKTTLYLQMNSLRPEDTAVYYCTIGGSLSVSSQGTLVTVSS (amino acids 397 to 511 of SEQ ID NO:76).

5. A pair of polypeptides according to any previous claim wherein said half-life extension domain has the sequence

EVQLVESGGGLVQPGNSLRRLSCAASGFTFSKFGMSWVRQAPGKGLEWVSSISGSGRDTLYAESV
KGRFTISRDNAAKTTLYLQMNSLRPEDTAVYYCTIGGSLSVSSQGTLVTVSSHHHHHH (amino acids 397 to 517 of SEQ ID NO:76).

6. A pair of polypeptides according to any previous claim wherein said first and said second target antigen binding domains bind a target antigen selected from the group consisting of EpCAM, EGFR and FolR.

7. A pair of polypeptides according to any previous claim wherein said first and said second protease cleavage site is cleaved by a human protease selected from the group consisting of MMP2, MMP9, meprin, matriptase, thrombin, uPA and cathepsin.

8. A pair of polypeptides according to any previous claim wherein said first and said second polypeptide each comprise an additional target antigen binding domain.
9. A pair of polypeptides according to any previous claim wherein said first and second target antigen binding domains are the same.
10. A pair of polypeptides according to any of claims 1 to 8 wherein said first and second target antigen binding domains are different.
11. A pair of polypeptides according to any previous claim wherein said aVH domain comprises the sequence
EVQLVESGGGLVQPGGSLKLSCAASGFTFNKYAMNWVRQAPGKGLEWVARIRSKYNNYATYY
ADSVKDRFTISRDDSKNTAYLQMNNLKTEDTAVYYCVRHGNFGNSYISYWAYWGQGTLVTVSS
(amino acids 137-261 of SEQ ID NO:44) and said aVL domain comprises
QTVVTQEPSLTVSPGGTVTLTCGSSTGAVTSGNYPNWVQQKPGQAPRGLIGGTKFLAPGTPARFS
GSLGGAALTLSGVQPEDEAEYYCVLWYSNRWVFGGGTKLTVL (amino acids 1-109 of SEQ
ID NO:46).
12. An expression vector comprising a first nucleic acid encoding said first polypeptide of any of claims 1 to 11 and a second nucleic acid encoding said second polypeptide of any of claims 1 to 11.
13. A host cell comprising the expression vector of claim 12.
14. Use of a pair of polypeptides according to any of claims 1 to 11 in the preparation of a medicament for the treatment of cancer.