Claims

1. A protein comprising at least one polypeptide chain having the formula

$$PP_1\text{-}PCL\text{-}PP_2,$$

wherein

PP_1 is a first polypeptide,

PP_2 is a second polypeptide, and

PCL is a protease-cleavable linker comprising at least one protease-cleavage site comprising the amino acid sequence $HRRX_1X_2RSVDE$ (SEQ ID NO: 42), wherein X_1 and X_2 are independently selected from the group consisting of KK, KQ, KR, QK, QQ, QR, RK, RQ, and RR.

2. The protein of claim 1, wherein the at least one protease-cleavage site comprises the amino acid sequence HRRRKRSVDE (SEQ ID NO: 43) or the amino acid sequence HRRQQRSVDE (SEQ ID NO: 44).

3. The protein of claim 1 or 2, wherein protease-cleavage of the linker results in a change in activity of the protein, wherein, preferably, the change in activity is binding or increased binding to at least one antigen.

4. The protein of any one of claims 1 to 3, wherein the protease-cleavable linker comprises two protease-cleavage sites, wherein the two protease-cleavage sites are located at the N-terminus and at the C-terminus of the protease-cleavable linker, respectively, wherein the two protease-cleavage sites may be the same or different.

5. The protein of any one of claims 1 to 4, wherein PP_1 comprises at least one immunoglobulin constant region and/or at least one immunoglobulin variable region, and/or wherein PP_2 comprises at least one immunoglobulin constant region and/or at least one immunoglobulin variable region, wherein, preferably,

PP_1 comprises an immunoglobulin constant region and PP_2 comprises an immunoglobulin variable region,

PP_1 comprises an immunoglobulin variable region and PP_2 comprises an immunoglobulin constant region,

PP_1 comprises an immunoglobulin constant region and PP_2 comprises an immunoglobulin constant region, or

PP_1 comprises an immunoglobulin variable region and PP_2 comprises an immunoglobulin variable region.

6. The protein of any one of claims 1 to 5, wherein the protein is an antibody or antibody derivative, wherein, preferably, the protein is a single chain antibody, preferably a multispecific single chain antibody.

7. The protein of any one of claims 1 to 6, comprising a polypeptide chain having a formula selected from the group consisting of:

(a) VL-CL-PCL-VH-CH1-hinge-CH2-CH3,

wherein

VL is an immunoglobulin light chain variable region;
CL is an immunoglobulin light chain constant region;
VH is an immunoglobulin heavy chain variable region;
CH1 is an immunoglobulin CH1 heavy chain constant region;
hinge is an immunoglobulin hinge region;
CH2 is an immunoglobulin CH2 heavy chain constant region;
CH3 is an immunoglobulin CH3 heavy chain constant region; and
PCL is the protease-cleavable linker;

(b) VL_a-CL_a-PCL-VL_b-CL_b,

wherein

VL_a is a first immunoglobulin light chain variable region;
VL_b is a second immunoglobulin light chain variable region;
CL_a is a first immunoglobulin light chain constant region;
CL_b is a second immunoglobulin light chain constant region; and
PCL is the protease-cleavable linker; and

(c) VL_a-CL_a-PCL-VL_b-CL_b-L-hinge-CH2-CH3

wherein

VL_a is a first immunoglobulin light chain variable region;
VL_b is a second immunoglobulin light chain variable region;
CL_a is a first immunoglobulin light chain constant region;
CL_b is a second immunoglobulin light chain constant region;

L is an optional linker;
hinge is an immunoglobulin hinge region;
CH2 is an immunoglobulin CH2 heavy chain constant region;
CH3 is an immunoglobulin CH3 heavy chain constant region; and
PCL is the protease-cleavable linker.

8. A nucleic acid or set of nucleic acids encoding the protein of any one of claims 1 to 7.

9. A vector or set of vectors comprising the nucleic acid or set of nucleic acids of claim 8.

10. A host cell comprising a protein of any one of claims 1 to 7, a nucleic acid or set of nucleic acids of claim 8, or a vector or set of vectors of claim 9, wherein, preferably, the host cell is a mammalian cell.

11. Method of producing a protein comprising the steps:

i) culturing a host cell according to claim 10; and
ii) isolating the protein from the host cell.

12. A protein obtainable by the method of claim 11.

13. The protein of any one of claims 1 to 7, or 12, wherein the protein is a therapeutically active protein.

14. The protein of claim 13 for use in therapy.