

CD38-expressing tumors still have a poor prognosis. Thus, there is a need for improved methods for treating such forms of cancer.

[0007] Thus while bispecifics generated from antibody fragments suffer biophysical and pharmacokinetic hurdles, a drawback of those built with full length antibody -like formats is that they engage co-target antigens multivalently in the absence of the primary target antigen, leading to nonspecific activation and potentially toxicity. The present invention solves this problem by introducing novel bispecific antibodies directed to CD3 and CD38.

BRIEF SUMMARY

[0008] In a first aspect, the present invention refers to a heterodimeric antibody comprising: a) a first monomer comprising: i) a first Fc domain; ii) an anti-CD3 scFv comprising SEQ ID NO: 18; wherein said scFv is covalently attached to the N-terminus of said Fc domain using a domain linker; b) a second monomer comprising a heavy chain comprising: i) a heavy variable domain; and ii) a heavy constant domain comprising a second Fc domain; and c) a light chain comprising a variable light domain and a constant light domain; wherein said heavy variable domain and said variable light domain bind CD38. In another aspect, the present invention refers to a nucleic acid composition encoding the heterodimeric antibody of the present invention, said composition comprising: a) a first nucleic acid encoding said first monomer; b) a second nucleic acid encoding said second monomer; and c) a third nucleic acid encoding said light chain. In yet another aspect, the present invention refers to an expression vector composition comprising: a) a first expression vector comprising a nucleic acid encoding said first monomer of the present invention; b) a second expression vector comprising a nucleic acid encoding said second monomer of the present invention; and c) a third expression vector comprising a nucleic acid encoding said light chain of the present invention. In still another aspect, the present invention refers to a host cell comprising the nucleic acid composition of the present invention. In another aspect, the present invention refers to a host cell comprising the expression vector composition of the present invention. In another aspect, the present invention refers to a method of making a heterodimeric antibody of the present invention comprising culturing the host cell of the present invention under conditions wherein said antibody is expressed, and recovering said

antibody. In yet another aspect, the present invention refers to the use of a heterodimeric antibody of the present invention in the preparation of a medicament for treating cancer in a patient in need thereof. In still another aspect, the present invention refers to a composition comprising a) a first expression vector comprising a nucleic acid encoding said first monomer of the present invention and b) a second expression vector encoding a nucleic acid encoding said second monomer of the present invention and a nucleic acid encoding said light chain of the present invention. Also described herein are heterodimeric antibodies directed against CD3 and CD38. In some examples, the heterodimeric antibodies comprise a first monomer comprising SEQ ID NO:91; a second monomer comprising SEQ ID NO:92; and a light chain comprising SEQ ID NO:93. In some examples, the heterodimeric antibodies comprise a first monomer comprising SEQ ID NO:88; a second monomer comprising SEQ ID NO:89; and a light chain comprising SEQ ID NO:90. The disclosure further provides nucleic acid compositions comprising first, second and third nucleic acids that encode the sequences above, as well as expression vectors comprising the nucleic acid compositions, host cells comprising either the nucleic acids or expression vectors, and methods of making and using the heterodimeric antibodies.

[0009] In an additional example, the disclosure provides heterodimeric antibodies comprising: a first monomer comprising: i) a first Fc domain; ii) an anti-CD3 scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached to the N-terminus of said Fc domain using a domain linker; a second monomer comprising a heavy chain comprising: i) a heavy variable domain; and ii) a heavy chain constant domain comprising a second Fc domain; and a light chain comprising a variable light domain and a variable light constant domain. In some examples the scFv variable light domain comprises: a vLCDR1 having SEQ ID NO:15, a vLCDR2 having SEQ ID NO:16 and a vLCDR3 having SEQ ID NO:17, said scFv variable heavy domain comprises a vhCDR1 having SEQ ID NO:11, a vhCDR2 having SEQ ID NO:12 and a vhCDR3 having SEQ

ID NO:13, and wherein said heavy variable domain and said variable light domain bind CD38.

[0010] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first Fc domain; ii) an anti-CD3 scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached to the N-terminus of said Fc domain using a domain linker; b) a second monomer comprising a heavy chain comprising: i) a heavy variable domain; and ii) a heavy chain constant domain comprising a second Fc domain; and c) a light chain comprising a variable light domain and a variable light constant domain. In this example, the scFv variable light domain comprises: a vlCDR1 having SEQ ID NO:24, a vlCDR2 having SEQ ID NO:25 and a vlCDR3 having SEQ ID NO:26, said scFv variable heavy domain comprises a vhCDR1 having SEQ ID NO:11, a vhCDR2 having SEQ ID NO:12 and a vhCDR3 having SEQ ID NO:13, and wherein said heavy variable domain and said variable light domain bind CD38.

[0011] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first Fc domain; ii) an anti-CD3 scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached to the N-terminus of said Fc domain using a domain linker; b) a second monomer comprising a heavy chain comprising: i) a heavy variable domain; and ii) a heavy chain constant domain comprising a second Fc domain; and c) a light chain comprising a variable light domain and a variable light constant domain. In this example, the scFv variable light domain comprises: a vlCDR1 having SEQ ID NO:33, a vlCDR2 having SEQ ID NO:34 and a vlCDR3 having SEQ ID NO:35, said scFv variable heavy domain comprises a vhCDR1 having SEQ ID NO:29, a vhCDR2 having SEQ ID NO:30 and a vhCDR3 having SEQ ID NO:31, and wherein said heavy variable domain and said variable light domain bind CD38.

[0012] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first Fc domain; ii) an anti-CD3 scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached to the N-terminus of said Fc domain using a domain linker; b) a second monomer comprising a heavy chain comprising: i) a heavy variable domain; and ii) a heavy chain constant domain comprising a second Fc domain; and c) a light chain comprising a

variable light domain and a variable light constant domain. In this example, the scFv variable light domain comprises: a vlCDR1 having SEQ ID NO:42, a vlCDR2 having SEQ ID NO:43 and a vlCDR3 having SEQ ID NO:44, said scFv variable heavy domain comprises a vhCDR1 having SEQ ID NO:38, a vhCDR2 having SEQ ID NO:39 and a vhCDR3 having SEQ ID NO:40, and wherein said heavy variable domain and said variable light domain bind CD38.

[0013] In an additional example, the “bottle opener” heterodimeric antibodies as described herein have a scFv that binds CD3 and vh and vl domains, wherein the variable light domain comprises: a vlCDR1 having the sequence RASQNVDTWVA (SEQ ID NO:69), a vlCDR2 having the sequence SASYRYS (SEQ ID NO:70) and a vlCDR3 having the sequence QQYDSYPLT (SEQ ID NO:71), said variable heavy domain comprises a vhCDR1 having the sequence RSWMN (SEQ ID NO:65), a vhCDR2 having the sequence EINPDSSTINYATSVKG (SEQ ID NO:66) and a vhCDR3 having the sequence YGNWFPY (SEQ ID NO:67).

[0014] In additional examples, the variable light domain comprises: a vlCDR1 having the sequence RASQNVDTNVA (SEQ ID NO:78), a vlCDR2 having the sequence SASYRYS (SEQ ID NO:79) and a vlCDR3 having the sequence QQYDSYPLT (SEQ ID NO:80), said variable heavy domain comprises a vhCDR1 having the sequence RSWMN (SEQ ID NO:74), a vhCDR2 having the sequence EINPDSSTINYATSVKG (SEQ ID NO:75) and a vhCDR3 having the sequence YGNWFPY (SEQ ID NO:76).

[0015] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first heavy chain comprising: 1) a first variable heavy domain; 2) a first constant heavy chain comprising a first Fc domain; 3) a scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached to the C-terminus of said Fc domain using a domain linker; b) a second monomer comprising a second heavy chain comprising a second variable heavy domain and a second constant heavy chain comprising a second Fc domain; and c) a common light chain comprising a variable light domain and a constant light domain; wherein said first and said second Fc domains have a set of amino acid substitutions selected from the group consisting of S364K/E357Q : L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K; T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q, and wherein said first variable heavy domain and said variable light domain

bind human CD38 (SEQ ID NO:131) , said second variable heavy domain and said variable light domain bind human CD38 (SEQ ID NO:131) , and said scFv binds human CD3 (SEQ ID NO:129).

[0016] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first heavy chain comprising: 1) a first variable heavy domain; 2) a first constant heavy domain comprising a first Fc domain; and 3) a first variable light domain, wherein said first variable light domain is covalently attached to the C-terminus of said first Fc domain using a domain linker; b) a second monomer comprising: i) a second variable heavy domain; ii) a second constant heavy domain comprising a second Fc domain; and iii) a third variable heavy domain, wherein said second variable heavy domain is covalently attached to the C-terminus of said second Fc domain using a domain linker; c) a common light chain comprising a variable light domain and a constant light domain; wherein said first and said second Fc domain have a set of amino acid substitutions selected from the group consisting of S364K/E357Q : L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K; T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q, wherein said first variable heavy domain and said variable light domain bind human CD38 (SEQ ID NO:131), said second variable heavy domain and said variable light domain bind said human CD38 (SEQ ID NO:131), and said second variable light domain and said third variable heavy domain binds human CD3 (SEQ ID NO:129).

[0017] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first heavy chain comprising: 1) a first variable heavy domain; 2) a first constant heavy chain comprising a first CH1 domain and a first Fc domain; 3) a scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached between the C-terminus of said CH1 domain and the N-terminus of said first Fc domain using domain linkers; b) a second monomer comprising a second heavy chain comprising a second variable heavy domain and a second constant heavy chain comprising a second Fc domain; and c) a common light chain comprising a variable light domain and a constant light domain; wherein said first and said second Fc domain have a set of amino acid substitutions selected from the group consisting of S364K/E357Q : L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K;

T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q, wherein said first variable heavy domain and said variable light domain bind human CD38 (SEQ ID NO:131), said second variable heavy domain and said variable light domain bind said human CD38 (SEQ ID NO:131), and said scFv binds human CD3 (SEQ ID NO:129).

[0018] In a further example, the disclosure provides heterodimeric antibodies comprising: a) a first monomer comprising: i) a first heavy chain comprising: 1) a first variable heavy domain; 2) a first constant heavy domain comprising a first Fc domain; and 3) a first variable light domain, wherein said second variable light domain is covalently attached between the C-terminus of the CH1 domain of said first constant heavy domain and the N-terminus of said first Fc domain using domain linkers; b) a second monomer comprising: i) a second variable heavy domain; ii) a second constant heavy domain comprising a second Fc domain; and iii) a third variable heavy domain, wherein said second variable heavy domain is covalently attached to the C-terminus of said second Fc domain using a domain linker; c) a common light chain comprising a variable light domain and a constant light domain; wherein said first and said second Fc domains have a set of amino acid substitutions selected from the group consisting of S364K/E357Q : L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K; T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q, wherein said first variable heavy domain and said variable light domain bind human CD38 (SEQ ID NO:131), said second variable heavy domain and said variable light domain bind said human CD38 (SEQ ID NO:131), and said second variable light domain and said third variable heavy domain binds human CD3 (SEQ ID NO:129).

[0019] In an additional example, the disclosure provides heterodimeric antibodies comprising a) a first monomer comprising: i) a first heavy chain comprising: 1) a first variable heavy domain; 2) a first constant heavy chain comprising a first CH1 domain and a first Fc domain; 3) a scFv comprising a scFv variable light domain, an scFv linker and a scFv variable heavy domain; wherein said scFv is covalently attached between the C-terminus of said CH1 domain and the N-terminus of said first Fc domain using domain linkers; b) a second monomer comprising a second Fc domain; and c) a light chain comprising a variable light domain and a constant light domain; wherein said first and said second Fc domain have a set of amino acid substitutions selected from the group consisting of S364K/E357Q :

L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K; T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q, wherein said first variable heavy domain and said variable light domain bind human CD38 (SEQ ID NO:131), said scFv binds human CD3 (SEQ ID NO:129).

[0020] In some examples the heterodimeric antibodies comprise a first Fc domain and a second Fc domain which comprise a set of variants selected from the group consisting of S364K/E357Q : L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K; T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q.

[0021] In further examples the scFv comprise scFv linkers that are charged linkers.

[0022] In additional examples the heavy chain constant domain of the heterodimeric antibodies outlined herein comprise the amino acid substitutions N208D/Q295E/N384D/Q418E/N421D.

[0023] In a further example, the heterodimeric antibodies of the disclosure have first and second Fc domains which comprise the amino acid substitutions E233P/L234V/L235A/G236del/S267K.

[0024] In an additional example, the disclosure provides nucleic acid composition encoding the heterodimeric antibodies as described herein that comprises a) a first nucleic acid encoding said first monomer; b) a second nucleic acid encoding said second monomer; and c) a third nucleic acid encoding said light chain.

[0025] In a further example, the disclosure provides expression vector compositions comprising: a) a first expression vector comprising a nucleic acid encoding said first monomer; b) a second expression vector comprising a nucleic acid encoding said second monomer; and c) a third expression vector comprising a nucleic acid encoding said light chain. The disclosure further provides host cells comprising either the nucleic acid compositions or the expression vector compositions.

[0026] The disclosure further provides methods of making the heterodimeric antibodies comprising culturing the host cells under conditions wherein said antibody is expressed, and recovering said antibody.

[0027] The disclosure further provides methods of treating cancer comprising administering a heterodimeric antibody as described herein to a patient in need thereof.

BRIEF DESCRIPTION OF THE DRAWINGS

[0028] Figures 1A and 1B depict several formats of the present invention. Two forms of the "bottle opener" format are depicted, one with the anti-CD3 antigen binding domain comprising a scFv and the anti-CD38 antigen binding domain comprising a Fab, and one with these reversed. The mAb-Fv, mAb-scFv, Central-scFv and Central-Fv formats are all shown. In addition, "one-armed" formats, where one monomer just comprises an Fc domain, are shown, both a one arm Central-scFv and a one arm Central-Fv. A dual scFv format is also shown.

[0029] Figure 2 depicts the sequences of the "High CD3" anti-CD3_H1.30_L1.47 construct, including the variable heavy and light domains (CDRs underlined), as well as the individual vl and vhCDRs, as well as an scFv construct with a charged linker (double underlined). As is true of all the sequences depicted in the Figures, this charged linker may be replaced by an uncharged linker or a different charged linker, as needed.

[0030] Figure 3 depicts the sequences of the "High-Int #1" Anti-CD3_H1.32_L1.47 construct, including the variable heavy and light domains (CDRs underlined), as well as the individual vl and vhCDRs, as well as an scFv construct with a charged linker (double underlined). As is true of all the sequences depicted in the Figures, this charged linker may be replaced by an uncharged linker or a different charged linker, as needed.

[0031] Figure 4 depicts the sequences of the "High-Int #2" Anti-CD3_H1.89_L1.47 construct, including the variable heavy and light domains (CDRs underlined), as well as the individual vl and vhCDRs, as well as an scFv construct with a charged linker (double underlined). As is true of all the sequences depicted in the Figures, this charged linker may be replaced by an uncharged linker or a different charged linker, as needed.

[0032] Figure 5 depicts the sequences of the "High-Int #3" Anti-CD3_H1.90_L1.47 construct, including the variable heavy and light domains (CDRs underlined), as well as the individual vl and vhCDRs, as well as an scFv construct with a charged linker (double underlined). As

WHAT IS CLAIMED IS:

1. A heterodimeric antibody comprising:

- a) a first monomer comprising:
 - i) a first Fc domain;
 - ii) an anti-CD3 scFv comprising SEQ ID NO: 18; wherein said scFv is covalently attached to the N-terminus of said Fc domain using a domain linker;
- b) a second monomer comprising a heavy chain comprising:
 - i) a heavy variable domain; and
 - ii) a heavy constant domain comprising a second Fc domain; and
- c) a light chain comprising a variable light domain and a constant light domain; wherein said heavy variable domain and said variable light domain bind CD38.

2. A heterodimeric antibody according to claim 1 wherein said variable light domain comprises: a vCDR1 having the sequence RASQNVDTWVA (SEQ ID NO:69), a vCDR2 having the sequence SASYRYS (SEQ ID NO:70) and a vCDR3 having the sequence QQYDSYPLT (SEQ ID NO:71), said variable heavy domain comprises a vhCDR1 having the sequence RSWMN (SEQ ID NO:65), a vhCDR2 having the sequence EINPDSSTINYATSVKG (SEQ ID NO:66) and a vhCDR3 having the sequence YGNWFPY (SEQ ID NO:67).

3. A heterodimeric antibody according to claim 1 wherein said variable light domain has the sequence DIVMTQSPSSLSASVGDRVTITCRASQNVDTWVAWYQQKPGQSPKALIYSASYRYS GVPD RFTGSGSGTDFTLTISSLQPEDFATYFCQQYDSYPLTFGGGTKLEIK (SEQ ID NO:68) and said variable heavy domain has the sequence EVQLVESGGGLVQPGGSLRLSCAASGFDFSRSWMNWVRQAPGKGLEWVSEINPDSSTIN YATSVKGRFTISRDN SKNTLYLQMNSLRAEDTAVYYCARYGNWFPYWGQGTLVTVSS (SEQ ID NO:64).

4. A heterodimeric antibody according to claim 1 wherein said variable light domain comprises: the vCDRs comprising a vCDR1 having the sequence RASQNVDTNVA (SEQ ID NO:78), a vCDR2 having the sequence SASYRYS (SEQ ID NO:79) and a vCDR3 having the sequence QQYDSYPLT (SEQ ID NO:80), said variable heavy domain comprises a vhCDR1 having the sequence RSWMN (SEQ ID NO:74), a vhCDR2 having the sequence

EINPDSSTINYATSVKG (SEQ ID NO:75) and a vhCDR3 having the sequence YGNWFPY (SEQ ID NO:76).

5. A heterodimeric antibody according to claim 1 wherein said variable light domain comprises the amino acid sequence DIVMTQSPSSLSASVGDRVTITCRASQNVDTNVAWYQQKPGQSPKALIYSASYRYS GVPDR FTGSGSGTDFTLTISLQPEDFATYFCQQYDSYPILTFGGGT and said variable heavy domain comprises the amino acid sequence EVQLVESGGGLVQPGGSLRLSCAASGFD FSRSWMNWVRQAPGKGLEWVSEINPDSSTIN YATSVKGRFTISRDN SKNTLYLQMNSLRAEDTAVYYCARYGNWFPYWGQGTLVTVSS.

6. A heterodimeric antibody according to claim 1 wherein said variable light domain comprises: the vCDRs comprising a vCDR1 having the sequence RASQNVDTWVA (SEQ ID NO:60), a vCDR2 having the sequence SASYRYS (SEQ ID NO:61) and a vCDR3 having the sequence QQYDSYPLT (SEQ ID NO:62), said variable heavy domain the vCDRs comprising a vhCDR1 having the sequence YSWMN (SEQ ID NO:56), a vhCDR2 having the sequence EINPQSSTINYATSVKG (SEQ ID NO:57) and a vhCDR3 having the sequence YGNWFPY (SEQ ID NO:58).

7. A heterodimeric antibody according to claim 1 wherein said variable light domain has the sequence SEQ ID NO:59 and said variable heavy domain has the sequence SEQ ID NO:55.

8. A heterodimeric antibody according to any of claims 1 to 7, wherein said first Fc domain and said second Fc domain comprise a set of variants selected from the group consisting of S364K/E357Q : L368D/K370S; L368D/K370S : S364K; L368E/K370S : S364K; T411T/E360E/Q362E : D401K; L368D/K370S : S364K/E357L and K370S : S364K/E357Q.

9. A heterodimeric antibody according to any of claims 1 to 8 wherein said heavy constant domain comprises the amino acid substitutions N208D/Q295E/N384D/Q418E/N421D.

10. A heterodimeric antibody according to any of claims 1 to 9 wherein said first and second Fc domains comprise the amino acid substitutions E233P/L234V/L235A/G236del/S267K.

11. A heterodimeric antibody according to claim 1, wherein the first monomer comprises SEQ ID NO:109; the second monomer comprises SEQ ID NO:104; and the light chain comprises SEQ ID NO:111.

12. A nucleic acid composition encoding the heterodimeric antibody of any of claims 1 to 11, said composition comprising:

- a) a first nucleic acid encoding said first monomer;
- b) a second nucleic acid encoding said second monomer; and
- c) a third nucleic acid encoding said light chain.

13. An expression vector composition comprising:

- a) a first expression vector comprising a nucleic acid encoding said first monomer of any of claims 1-11;
- b) a second expression vector comprising a nucleic acid encoding said second monomer of any of claims 1-11; and
- c) a third expression vector comprising a nucleic acid encoding said light chain of any of claims 1-11.

14. A host cell comprising the nucleic acid composition of claim 12.

15. A host cell comprising the expression vector composition of claim 13.

16. A method of making a heterodimeric antibody according to any of claims 1 to 11 comprising culturing the host cell of claim 14 or 15 under conditions wherein said antibody is expressed, and recovering said antibody.

17. Use of a heterodimeric antibody according to any of claims 1 to 11 in the preparation of a medicament for treating cancer in a patient in need thereof.

18. A composition comprising a) a first expression vector comprising a nucleic acid encoding said first monomer of any one of claims 1-11 and b) a second expression vector encoding a nucleic acid encoding said second monomer of any one of claims 1-11 and a nucleic acid encoding said light chain of any one of claims 1-11.