

employed in cancer treatment such as, for example, chemotherapeutic agents, non-chemotherapeutic agents (e.g. anti-PD-1 or anti-PD-L1 inhibitors, such as antagonist antibodies), anti-neoplastic agents, and/or radiation. For example, administration can occur before, during, and/or after any of the treatments described herein. Examples of chemotherapeutic agents are discussed herein and include, but are not limited to, cisplatin, taxol, etoposide, mitoxantrone (Novantrone®), actinomycin D, cycloheximide, camptothecin (or water soluble derivatives thereof), methotrexate, mitomycin (e.g., mitomycin C), dacarbazine (DTIC), anti-neoplastic antibiotics such as adriamycin (doxorubicin) and daunomycin, and all the chemotherapeutic agents mentioned herein.

[0069] In some examples, a TCE molecule of the present disclosure may be administered concurrently with, before, or after a PD-1 antagonist antibody or a PD-L1 antagonist antibody. The term “PD-1 antagonist antibody” refers to an antibody that specifically binds to PD-1 and decreases, blocks, inhibits, abrogates or interferes with signal transduction resulting from the interaction of PD-1 and one or more of its ligands, such as PD-L1 and PD-L2. In some examples, a PD-1 antagonist antibody inhibits the binding of PD-1 to PD-L1 and/or PD-L2. The term “PD-L1 antagonist antibody” refers to an antibody that specifically binds to PD-L1 and decreases, blocks, inhibits, abrogates or interferes with signal transduction resulting from the interaction of PD-L1 with the PD-1 receptor. In some examples, a PD-L1 antagonist antibody inhibits the binding of PD-L1 to PD-1. In some examples, the PD-1 antagonist is any one of Antibody 20C1.006 (SEQ ID NOs: 179-188), Antibody 20C1.009 (SEQ ID NOs: 139-148, or 139-147 and 212), Antibody 20A2.3 (SEQ ID NOs: 149-158), Antibody 20D4.6 (SEQ ID NOs: 159-168), or Antibody 20D4.17 (SEQ ID NOs: 169-178). In one example, the PD-1 antagonist antibody is pembrolizumab. In another example, the PD-1 antagonist antibody is nivolumab. In yet another example, the PD-1 antagonist antibody is cemiplimab. In a particular example, the PD-1 antagonist antibody is antibody 20C1.009, for which the amino acid sequences of the CDRs, variable regions, and full light and heavy chains are provided in SEQ ID NOs: 139-148 and 212. 20C1.009 is also known as AMG 404 and is also known as zelumab. In exemplary aspects, an anti-PD-1 antibody such as 20C1.009 comprises a HC comprising a C-terminal lysine, as in SEQ ID NO: 148. In alternative aspects, the antibody comprises a HC without the C-terminal lysine, as in SEQ ID NO: 212.

[0070] In some examples, the 1185-1200

[0070a] In one aspect, the present disclosure refers to a T cell engager (TCE) molecule comprising (i) an scFab that binds to a tumor antigen, wherein the scFab comprises a first heavy chain variable region (scFab VH), a CH1 domain, a first light chain variable region (scFab VL), a C κ or C λ domain, and a linker, wherein the linker is (G4S)₆, (G4S)₇, (G4S)₈, (G4Q)₆, (G4Q)₇, or (G4Q)₈, (ii) an scFv that binds CD3, comprising a second VL and a second VH, and (iii) an scFc, and wherein the TCE molecule is a single chain.

[0070b] In another aspect, the present disclosure refers to use of the TCE molecule as disclosed herein in the manufacture of a medicament for treating cancer.

[0070c] In another aspect, the present disclosure refers to a pharmaceutical composition comprising the TCE molecule as disclosed herein and one or more pharmaceutically acceptable carriers, diluents, or excipients.

BRIEF DESCRIPTION OF THE DRAWINGS

CLAIMS

We claim:

1. A T cell engager (TCE) molecule comprising (i) an scFab that binds to a tumor antigen, wherein the scFab comprises a first heavy chain variable region (scFab VH), a CH1 domain, a first light chain variable region (scFab VL), a C κ or C λ domain, and a linker, wherein the linker is (G4S)₆, (G4S)₇, (G4S)₈, (G4Q)₆, (G4Q)₇, or (G4Q)₈, (ii) an scFv that binds CD3, comprising a second VL and a second VH, and (iii) an scFc, and wherein the TCE molecule is a single chain.
2. The TCE molecule of Claim 1, wherein the scFab comprises a C-terminus portion that is connected by a linker to an N-terminal portion of the scFv.
3. The TCE molecule of Claim 1, wherein the scFc comprises an N-terminus portion that is connected by a linker to a C-terminal portion of the scFv.
4. The TCE molecule of any one of Claims 1-3, wherein the scFv binds human CD3.
5. The TCE molecule of any one of Claims 1-4, wherein the scFab has an orientation in the following order, from N-terminus to C-terminus, VH, CH1, VL, and either C κ or C λ .
6. The TCE molecule of any one of Claims 1-4, wherein the scFab has an orientation in the following order, from N-terminus to C-terminus, VL, either C κ or C λ , VH, and CH1.
7. The TCE molecule of any one of Claims 1-5, wherein the scFab comprises a linker that connects the CH1 and scFab VL, wherein the linker is (G4S)₆, (G4S)₇, (G4S)₈, (G4Q)₆, (G4Q)₇, or (G4Q)₈.
8. The TCE molecule of any one of Claims 1-4 and 6, wherein the TCE molecule comprises a linker that connects the scFab C κ or C λ and the scFab VH, wherein the linker is (G4S)₆, (G4S)₇, (G4S)₈, (G4Q)₆, (G4Q)₇, or (G4Q)₈.
9. The TCE molecule of any one of Claims 1-8, wherein CH1, C κ and/or C λ domains are IgG, IgM, IgA, IgD, or IgE.
10. The TCE molecule of Claim 9, wherein the domains are IgG.
11. The TCE molecule of Claim 9 or 10, wherein the domains are IgG1.
12. The TCE molecule of any one of Claims 1-11, wherein the scFab contains a cysteine clamp between CH1 and either C κ or C λ .
13. The TCE molecule of any one of Claims 1-12, wherein the orientation of the TCE molecule from N-terminus to C-terminus is: VH-CH1-Linker-VL-C κ or C λ -

Linker-VH-Linker-VL-Linker- Fc1 (CH2-CH3) -Linker-Fc2 (CH2-CH3) and wherein the TCE molecule is a single chain.

14. The TCE molecule of any one of Claims 1-12, wherein the orientation of the TCE molecule from N-terminus to C-terminus is: VL-CH1-Linker-VH-Ck or Cλ-Linker-VH-Linker-VL-Linker- Fc1 (CH2-CH3) -Linker-Fc2 (CH2-CH3) and wherein the TCE molecule is a single chain.
15. The TCE molecule of any one of Claims 1-14, wherein the tumor antigen is CCR8.
16. The TCE molecule of any one of Claims 1-15, wherein the scFv that binds CD3 comprises HCDR1 comprising an amino acid sequence given by SEQ ID NO: 199, HCDR2 comprising an amino acid sequence given by SEQ ID NO: 200, HCDR3 comprising an amino acid sequence given by SEQ ID NO: 201, LCDR1 comprising an amino acid sequence given by SEQ ID NO: 202, LCDR2 comprising an amino acid sequence given by SEQ ID NO: 203, and LCDR3 comprising an amino acid sequence given by SEQ ID NO: 204. .
17. The TCE molecule of any one of Claims 1-15, wherein the scFv that binds CD3 comprises HCDR1 comprising an amino acid sequence given by SEQ ID NO: 191, HCDR2 comprising an amino acid sequence given by SEQ ID NO: 192, HCDR3 comprising an amino acid sequence given by SEQ ID NO: 193, LCDR1 comprising an amino acid sequence given by SEQ ID NO: 194, LCDR2 comprising an amino acid sequence given by SEQ ID NO: 195, and LCDR3 comprising an amino acid sequence given by SEQ ID NO: 196.
18. The TCE molecule of any one of Claims 1-17 for use in therapy.
19. Use of the TCE molecule of any one of Claims 1-17 in the manufacture of a medicament for treating cancer.
20. The use of Claim 19, wherein the cancer is a solid tumor.
21. The use of Claim 19 or 20, wherein the cancer is non-small cell lung cancer, gastric cancer, head and neck squamous cell carcinoma, hepatocellular carcinoma, triple-negative breast cancer, colorectal cancer, pancreatic cancer, or metastatic castrate-resistant prostate cancer.
22. The use of Claims 19-21, wherein the cancer is non-small cell lung cancer, gastric cancer, head and neck squamous cell carcinoma, hepatocellular carcinoma, or triple-negative breast cancer.

23. The use of Claims 19-22, wherein the medicament is to be administered with a PD-1 antagonist antibody or PD-L1 antagonist antibody.
24. The use of Claim 23, wherein the PD-1 antagonist antibody or PD-L1 antagonist antibody is to be administered prior to, concurrently with, and/or after administration of the TCE molecule.
25. The use of Claim 23 or 24, wherein the PD-1 antagonist antibody is pembrolizumab, nivolumab, cemiplimab, or an antibody that comprises HCDR1 comprising an amino acid sequence given by SEQ ID NO: 142, HCDR2 comprising an amino acid sequence given by SEQ ID NO: 143, HCDR3 comprising an amino acid sequence given by SEQ ID NO: 144, LCDR1 comprising an amino acid sequence given by SEQ ID NO: 139, LCDR2 comprising an amino acid sequence given by SEQ ID NO: 140, and LCDR3 comprising an amino acid sequence given by SEQ ID NO: 141..
26. The use of Claim 23 or 24, wherein the PD-L1 antagonist antibody is atezolizumab, avelumab, or durvalumab.
27. A pharmaceutical composition comprising the TCE molecule of any one of Claims 1-17 and one or more pharmaceutically acceptable carriers, diluents, or excipients.