

tumor cells, resulted in more effective control of mouse tumor growth than targeting either pathway alone (Sakuishi et. al. JEM 2010, 207: 2187 and WO 2015/048312).

An oral, small molecule that antagonizes PD-L1 and TIM-3 has been reported (Sasikumar et al., AACR; Cancer Res 2016;76(14 Suppl):Abstract 4861). This small
5 molecule compound was reported to demonstrate efficacy in syngenic mouse tumor models.

There remains a need to provide compounds that can bind and inhibit both human PD-L1 and human TIM-3, but also have the target specificity and pharmacokinetics of an antibody. In particular, there remains a need to provide heterodimeric bispecific
10 antibodies that bind both human PD-L1 and human TIM-3 where the heterodimeric bispecific antibody enables pairing of two different heavy chains and two different light chains into a single IgG-like antibody. Further, there is a need for the bispecific antibody to resemble native antibody structure as measured by modeling, mass spectrometry, and stability analyses for purposes such as low immunogenicity, stable *in vivo*
15 pharmacokinetics, and adequate stability. Additionally, there remains a need to provide anti-human PD-L1 and anti-human TIM-3 bispecific antibodies that possess one or more of the following features: simultaneously bind and bridge PD-L1 expressing tumor cells and TIM-3 expressing T-cells, suppress PD-1 and TIM-3 checkpoint inhibitor pathways, exhibit better anti-tumor activity as measured in a tumor model than a combination of an
20 anti-PD-L1 antibody and an anti-TIM-3 antibody, and lack antibody immune effector functions.

Summary

In an aspect of the present disclosure, there is provided an antibody that binds human PD-
25 L1 (SEQ ID NO: 1) and human TIM-3 (SEQ ID NO: 2) comprising:

- a) a first heavy chain (HC) comprising the amino acid sequences of SEQ ID NO: 3, SEQ ID NO: 19, and SEQ ID NO: 22,
- b) a first light chain (LC) comprising the amino acid sequences of SEQ ID NO: 4 and SEQ ID NO: 27,
- 30 c) a second HC comprising the amino acid sequences of SEQ ID NO: 7, SEQ ID NO: 23, and SEQ ID NO: 26,

- d) a second LC comprising the amino acid sequences of SEQ ID NO: 10 and SEQ ID NO: 28;

and wherein, the first LC forms an inter-chain disulfide bond with the first HC, the second LC forms an inter-chain disulfide bond with the second HC, and the first HC forms two inter-chain disulfide bonds with the second HC, and the first HC and second HC are human IgG1 isotype.

In another aspect of the present disclosure, there is provided an antibody that binds human PD-L1 (SEQ ID NO: 1) and human TIM-3 (SEQ ID NO: 2) comprising two light chains (LC) and two heavy chains (HC), wherein:

- 10 a) the first LC has the amino acid sequence of SEQ ID NO: 12,
b) the first HC has the amino acid sequence of SEQ ID NO: 11,
c) the second LC has the amino acid sequence of SEQ ID NO: 16, SEQ ID NO: 17, or SEQ ID NO: 18, and
d) the second HC has the amino acid sequence of SEQ ID NO: 13, SEQ ID
15 NO: 14, or SEQ ID NO: 15.

In a further aspect of the present disclosure, there is provided a mammalian cell comprising a DNA molecule comprising a polynucleotide sequence encoding polypeptides having the amino acid sequences of SEQ ID NO: 11, SEQ ID NO: 15, SEQ ID NO: 12, and SEQ ID NO: 18, wherein the cell is capable of expressing the antibody
20 disclosed herein.

In another aspect of the present disclosure, there is provided a mammalian cell comprising a first DNA molecule and a second DNA molecule, wherein the first DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 11 and SEQ ID NO: 12, and wherein the second DNA
25 molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 15 and SEQ ID NO: 18, wherein the cell is capable of expressing the antibody disclosed herein.

In a further aspect of the present disclosure, there is provided a process for producing an antibody comprising cultivating the mammalian cell disclosed herein under
30 conditions such that the antibody is expressed, and recovering the expressed antibody.

In another aspect of the present disclosure, there is provided an antibody produced by cultivating a mammalian cell comprising a DNA molecule comprising a

polynucleotide sequence encoding polypeptides having the amino acid sequences of SEQ ID NO: 11, SEQ ID NO: 15, SEQ ID NO: 12, and SEQ ID NO: 18 under conditions such that the antibody is expressed, and recovering the expressed antibody.

5 In a further aspect of the present disclosure, there is provided an antibody produced by cultivating a mammalian cell comprising a first DNA molecule and a second DNA molecule, wherein the first DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 11 and SEQ ID NO: 12, and wherein the second DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 15 and SEQ ID
10 NO: 18 under conditions such that the antibody is expressed, and recovering the expressed antibody.

In another aspect of the present disclosure, there is provided a pharmaceutical composition, comprising the antibody disclosed herein and an acceptable carrier, diluent, or excipient.

15 In a further aspect of the present disclosure, there is provided a use of an antibody disclosed herein, in the manufacture of a medicament for treating cancer.

In another aspect of the present disclosure, there is provided an antibody disclosed herein, for use in therapy.

In a further aspect of the present disclosure, there is provided an antibody disclosed
20 herein, for use in the treatment of cancer.

In another aspect of the present disclosure, there is provided a use of an antibody disclosed herein, in the manufacture of a medicament for treating cancer, wherein the antibody is to be administered in simultaneous, separate, or sequential combination with one or more standard of care agents selected from the group consisting of cisplatin,
25 carboplatin, dacarbazine, liposomal doxorubicin, docetaxel, cyclophosphamide and doxorubicin, navelbine, eribulin, paclitaxel, paclitaxel protein-bound particles for injectable suspension, ixabepilone, capecitabine, FOLFOX (leucovorin, fluorouracil, and oxaliplatin), FOLFIRI (leucovorin, fluorouracil, and irinotecan), gemcitabine, topotecan, liposomal irinotecan, pemetrexed, and cetuximab.

30 In a further aspect of the present disclosure, there is provided a pharmaceutical composition for use in treating cancer, comprising an effective amount of the antibody disclosed herein.

Description

Accordingly, the present invention provides an antibody that binds human PD-L1 (SEQ ID NO: 1) and human TIM-3 (SEQ ID NO: 2) comprising:

- 5 a) a first heavy chain (HC) comprising a heavy chain variable region (HCVR) having the amino acid sequence of SEQ ID NO: 3,
- b) a first light chain (LC) comprising a light chain variable region (LCVR) having the amino acid sequence of SEQ ID NO: 4,
- c) a second HC comprising a HCVR having the amino acid sequence of SEQ ID
10 NO: 5, SEQ ID NO: 6, or SEQ ID NO: 7,
- d) a second LC comprising a LCVR having the amino acid sequence of SEQ ID
 NO: 8, SEQ ID NO: 9, or SEQ ID NO: 10;

WE CLAIM:

1. An antibody that binds human PD-L1 (SEQ ID NO: 1) and human TIM-3 (SEQ ID NO: 2) comprising:
 - 5 a) a first heavy chain (HC) comprising the amino acid sequences of SEQ ID NO: 3, SEQ ID NO: 19, and SEQ ID NO: 22,
 - b) a first light chain (LC) comprising the amino acid sequences of SEQ ID NO: 4 and SEQ ID NO: 27,
 - c) a second HC comprising the amino acid sequences of SEQ ID NO: 7, SEQ ID NO: 23, and SEQ ID NO: 26,
 - 10 d) a second LC comprising the amino acid sequences of SEQ ID NO: 10 and SEQ ID NO: 28;and wherein, the first LC forms an inter-chain disulfide bond with the first HC, the second LC forms an inter-chain disulfide bond with the second HC, and the first HC forms two inter-chain disulfide bonds with the
15 second HC, and the first HC and second HC are human IgG1 isotype.
2. The antibody of Claim 1, wherein:
 - a) the first HC comprises the amino acid sequences of SEQ ID NO: 20 and SEQ ID NO: 21, and
 - 20 b) the second HC comprises the amino acid sequences of SEQ ID NO: 24 and SEQ ID NO: 25.
3. An antibody that binds human PD-L1 (SEQ ID NO: 1) and human TIM-3 (SEQ ID NO: 2) comprising two light chains (LC) and two heavy chains (HC), wherein:
 - a) the first LC has the amino acid sequence of SEQ ID NO: 12,
 - 25 b) the first HC has the amino acid sequence of SEQ ID NO: 11,
 - c) the second LC has the amino acid sequence of SEQ ID NO: 16, SEQ ID NO: 17, or SEQ ID NO: 18, and
 - d) the second HC has the amino acid sequence of SEQ ID NO: 13, SEQ ID NO: 14, or SEQ ID NO: 15.
- 30 4. The antibody of Claim 3, wherein the second LC has the amino acid sequence of SEQ ID NO: 16, and the second HC has the amino acid sequence of SEQ ID NO: 13.

5. The antibody of Claim 3, wherein the second LC has the amino acid sequence of SEQ ID NO: 17, and the second HC has the amino acid sequence of SEQ ID NO: 14.
6. The antibody of Claim 3, wherein the second LC has the amino acid sequence of SEQ ID NO: 18, and the second HC has the amino acid sequence of SEQ ID NO: 15.
7. A mammalian cell comprising a DNA molecule comprising a polynucleotide sequence encoding polypeptides having the amino acid sequences of SEQ ID NO: 11, SEQ ID NO: 15, SEQ ID NO: 12, and SEQ ID NO: 18, wherein the cell is capable of expressing the antibody of Claim 6.
8. A mammalian cell comprising a first DNA molecule and a second DNA molecule, wherein the first DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 11 and SEQ ID NO: 12, and wherein the second DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 15 and SEQ ID NO: 18, wherein the cell is capable of expressing the antibody of Claim 6.
9. A process for producing an antibody comprising cultivating the mammalian cell of Claim 7 or 8 under conditions such that the antibody is expressed, and recovering the expressed antibody.
10. An antibody produced by cultivating a mammalian cell comprising a DNA molecule comprising a polynucleotide sequence encoding polypeptides having the amino acid sequences of SEQ ID NO: 11, SEQ ID NO: 15, SEQ ID NO: 12, and SEQ ID NO: 18 under conditions such that the antibody is expressed, and recovering the expressed antibody.
11. An antibody produced by cultivating a mammalian cell comprising a first DNA molecule and a second DNA molecule, wherein the first DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 11 and SEQ ID NO: 12, and wherein the second DNA molecule comprises a polynucleotide sequence encoding polypeptides having the amino acid sequence of SEQ ID NO: 15 and SEQ ID NO: 18 under conditions such that the antibody is expressed, and recovering the expressed antibody.

12. A pharmaceutical composition, comprising the antibody of any one of Claims 1-6, or 10-11 and an acceptable carrier, diluent, or excipient.
13. Use of an antibody of any one of claims 1-6, or 10-11, in the manufacture of a medicament for treating cancer.
- 5 14. The use of Claim 13, wherein the cancer is melanoma, lung cancer, head and neck cancer, liver cancer, colorectal cancer, pancreatic cancer, gastric cancer, kidney cancer, bladder cancer, prostate cancer, breast cancer, ovarian cancer, endometrial cancer, esophageal cancer, soft tissue sarcoma, cholangiocarcinoma, or hepatocellular carcinoma.
- 10 15. The use of Claim 14, wherein the lung cancer is non-small cell lung cancer, small cell lung cancer, or mesothelioma.
- 15 16. The use of any one of Claims 13-15, wherein one or more standard of care agents selected from the group consisting of cisplatin, carboplatin, dacarbazine, liposomal doxorubicin, docetaxel, cyclophosphamide and doxorubicin, navelbine, eribulin, paclitaxel, paclitaxel protein-bound particles for injectable suspension, ixabepilone, capecitabine, FOLFOX (leucovorin, fluorouracil, and oxaliplatin), FOLFIRI (leucovorin, fluorouracil, and irinotecan), gemcitabine, topotecan, liposomal irinotecan, pemetrexed, and cetuximab are to be further administered simultaneously, separately, or sequentially.
- 20 17. The antibody of any one of Claims 1-6, or 10-11 for use in therapy.
18. The antibody of any one of Claims 1-6, or 10-11 for use in the treatment of cancer.
- 25 19. The antibody for use of Claim 18, wherein the cancer is melanoma, lung cancer, head and neck cancer, liver cancer, colorectal cancer, pancreatic cancer, gastric cancer, kidney cancer, bladder cancer, prostate cancer, breast cancer, ovarian cancer, endometrial cancer, esophageal cancer, soft tissue sarcoma, cholangiocarcinoma, or hepatocellular carcinoma.
20. The antibody for use of Claim 19, wherein the lung cancer is non-small cell lung cancer, small cell lung cancer, or mesothelioma.
- 30 21. Use of an antibody of any one of Claims 1-6, or 10-11, in the manufacture of a medicament for treating cancer, wherein the antibody is to be administered in simultaneous, separate, or sequential combination with one or more standard of

- care agents selected from the group consisting of cisplatin, carboplatin, dacarbazine, liposomal doxorubicin, docetaxel, cyclophosphamide and doxorubicin, navelbine, eribulin, paclitaxel, paclitaxel protein-bound particles for injectable suspension, ixabepilone, capecitabine, FOLFOX (leucovorin, 5 fluorouracil, and oxaliplatin), FOLFIRI (leucovorin, fluorouracil, and irinotecan), gemcitabine, topotecan, liposomal irinotecan, pemetrexed, and cetuximab.
22. A pharmaceutical composition for use in treating cancer, comprising an effective amount of the antibody of any one of claims 1-6, or 10-11.
23. The composition for use of Claim 22, wherein the cancer is melanoma, lung 10 cancer, head and neck cancer, liver cancer, colorectal cancer, pancreatic cancer, gastric cancer, kidney cancer, bladder cancer, prostate cancer, breast cancer, ovarian cancer, endometrial cancer, esophageal cancer, soft tissue sarcoma, cholangiocarcinoma, or hepatocellular carcinoma.
24. The composition for use of Claim 23, wherein the lung cancer is non-small cell 15 lung cancer, small cell lung cancer, or mesothelioma.
25. The composition for use of any one of Claims 22-24, which is to be administered in simultaneous, separate, or sequential combination with one or more standard of care agents selected from the group consisting of cisplatin, carboplatin, 20 dacarbazine, liposomal doxorubicin, docetaxel, cyclophosphamide and doxorubicin, navelbine, eribulin, paclitaxel, paclitaxel protein-bound particles for injectable suspension, ixabepilone, capecitabine, FOLFOX (leucovorin, fluorouracil, and oxaliplatin), FOLFIRI (leucovorin, fluorouracil, and irinotecan), gemcitabine, topotecan, liposomal irinotecan, pemetrexed, and cetuximab.
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