

~~Hence there is the need to generate and/or select further improved, truly HLA-G specific antibodies with receptor inhibition properties.~~

Summary

The disclosure provides an antibody that bind to human HLA-G (and that inhibits ILT2 binding to HLAG on JEG-3 cells (ATCC HTB36) and restores HLA-G specific suppressed TNF alpha release by monocytes co-cultured with JEG-3 cells. ~~In one aspect, the present disclosure provides an isolated antibody that bind to human HLA-G; and that inhibits ILT2 binding to HLAG on JEG-3 cells (ATCC HTB36) and restores HLA-G specific suppressed TNF alpha release by monocytes co-cultured with JEG-3 cells~~

In ~~another aspect~~ one example, the present disclosure provides an isolated antibody that binds to human HLA-G (in one embodiment example the antibody binds to HLA-G β 2M MHC I complex comprising SEQ ID NO: 43), wherein the antibody comprises

- A) (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:1, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:2, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:3; and (b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:4; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:5 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:6; or
- B) (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:9, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:10, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:11; and (b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:12; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:13 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:14; or
- C) (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:17, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:18, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:19; and (b) a VL domain comprising (i) HVR-L1 comprising the

amino acid sequence of SEQ ID NO:28; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:29 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:30.

Thus, in one aspect, the present disclosure provides an isolated antibody that bind to human HLA-G; and that inhibits ILT2 binding to HLA-G on JEG-3 cells (ATCC HTB36) and restores HLA-G specific suppressed TNF alpha release by monocytes co-cultured with JEG-3 cells, wherein the antibody comprises

(a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:1, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:2, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:3; and

(b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:4; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:5 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:6.

One example of the disclosure is an isolated antibody that binds to human HLA-G(in one ~~embodiment~~ example the antibody binds to HLA-G β 2M MHC I complex comprising SEQ ID NO: 43), wherein the antibody

A)

- i) comprises a VH sequence of SEQ ID NO:7 and a VL sequence of SEQ ID NO:8;
- ii) or humanized variant of the VH and VL of the antibody under i); or

B)

- i) comprises a VH sequence of SEQ ID NO:15 and a VL sequence of SEQ ID NO:16;
- ii) or humanized variant of the VH and VL of the antibody under i); or

C)

i) comprises a VH sequence of SEQ ID NO:23 and a VL sequence of SEQ ID NO:24;

5 ii) or humanized variant of the VH and VL of the antibody under i); or

D)

i) comprises a VH sequence of SEQ ID NO:31 and a VL sequence of SEQ ID NO:32;

ii) or humanized variant of the VH and VL of the antibody under i).

10 In another aspect, the present disclosure provides an isolated antibody that binds to human HLA-G, wherein the antibody

a) binds to the same epitope as an antibody which comprises a VH sequence of SEQ ID NO:7 and a VL sequence of SEQ ID NO:8;

15 or b) binds to the same epitope as an antibody which comprises a VH sequence of SEQ ID NO:31 and a VL sequence of SEQ ID NO:32.

In one example the anti-HLA-G antibody described herein

a) does not crossreact with a modified human HLA-G β 2M MHC I complex comprising SEQ ID NO:44; and/ or

~~In another aspect, the~~The present disclosure provides the use of the antibody described herein in the manufacture of a medicament. In one example the medicament is for treatment of cancer. Thus, in one aspect, the present disclosure provides use of the antibody as described herein in the manufacture of a medicament for treating cancer.

The disclosure provides a method of treating an individual having cancer comprising administering to the individual an effective amount of the antibody described herein.

The present disclosure uses tailor-made chimeric antigens and/or stringent screening assays to identify HLA-G specific antibodies among numerous candidates (avoiding cross-reactivity to other MHC class I complex molecules and at the same time

Patent Claims

1. An isolated antibody that bind to human HLA-G; and that inhibits ILT2 binding to HLA-G on JEG-3 cells (ATCC HTB36) and restores HLA-G specific suppressed TNF alpha release by monocytes co-cultured with JEG-3 cells, wherein the antibody comprises

(a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:1, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:2, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:3; and

(b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:4; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:5 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:6.

- ~~2. An isolated antibody that bind to human HLA-G wherein the antibody comprises~~

~~A) (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:1, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:2, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:3; and (b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:4; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:5 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:6; or~~

~~B) (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:9, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:10, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:11; and (b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:12; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:13 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:14; or~~

~~C) (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:17, (ii) HVR-H2 comprising the amino acid sequence of SEQ~~

5 ID NO:18, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:19; and (b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:20; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:21 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:22; or

10 D) — (a) a VH domain comprising (i) HVR-H1 comprising the amino acid sequence of SEQ ID NO:25, (ii) HVR-H2 comprising the amino acid sequence of SEQ ID NO:26, and (iii) HVR-H3 comprising an amino acid sequence selected from SEQ ID NO:27; and (b) a VL domain comprising (i) HVR-L1 comprising the amino acid sequence of SEQ ID NO:28; (ii) HVR-L2 comprising the amino acid sequence of SEQ ID NO:29 and (iii) HVR-L3 comprising the amino acid sequence of SEQ ID NO:30.

32. The antibody according to claim 21, wherein the antibody comprises

A)

15 ~~iv~~i) ~~comprises~~ a VH sequence of SEQ ID NO:7 and a VL sequence of SEQ ID NO:8; or

~~v~~ii) ~~or~~ humanized variant of the VH and VL of the antibody under i); or

~~viii~~iii) ~~comprises~~ a VH sequence of SEQ ID NO:33 and a VL sequence of SEQ ID NO:34; ~~or~~

20 B)

~~comprises a VH sequence of SEQ ID NO:15 and a VL sequence of SEQ ID NO:16; or~~

C)

25 ~~comprises a VH sequence of SEQ ID NO:23 and a VL sequence of SEQ ID NO:24; or~~

D)

~~comprises a VH sequence of SEQ ID NO:31 and a VL sequence of SEQ ID NO:32.~~

43. An isolated antibody that binds to human HLA-G, wherein the antibody
- a) binds to the same epitope as an antibody which comprises a VH sequence of SEQ ID NO:7 and a VL sequence of SEQ ID NO:8;
- or b) binds to the same epitope as an antibody which comprises a VH sequence of SEQ ID NO:31 and a VL sequence of SEQ ID NO:32.
54. The anti-HLA-G antibody according to any one of claims 1 to 43, wherein the antibody
- a) does not crossreact with a modified human HLA-G β 2M MHC I complex comprising SEQ ID NO:44; and/ or
- b) does not crossreact with human HLA-A2 β 2M MHC I complex comprising SEQ ID NO:39 and SEQ ID NO: 37; and/ or
- c) does not crossreact with a mouse H2Kd β 2M MHC I complex comprising SEQ ID NO:45; and/ or
- d) does not crossreact with rat RT1A β 2M MHC I complex comprising SEQ ID NO:47; and/ or
- e) inhibits ILT2 binding to monomeric HLA-G β 2M MHC I complex; and/or
- f) inhibits ILT2 binding to trimeric HLA-G β 2M MHC I complex; and/or
- g) inhibits ILT2 binding to monomeric and/or dimeric and/or trimeric HLA-G β 2M MHC I complex by more than 50%; and/ or
- ~~h) inhibits ILT2 binding to JEG3 cells (ATCC No. HTB36); and/or~~
- ~~i) binds to (HLA-G on) JEG3 cells (ATCC No. HTB36), and inhibits ILT2 binding to (HLA-G on) JEG3 cells (ATCC No. HTB36); and/or~~
- ~~j) inhibits CD8a binding to HLA-G by more than 80%.~~
65. The antibody according to any one of the preceding claims, wherein the antibody is of IgG1 isotype.

~~76~~. The antibody according claim ~~65~~, wherein the antibody is of IgG1 isotype with mutations L234A, L235A and P329G (numbering according to the EU index of Kabat).

5 ~~87~~. Isolated nucleic acid encoding the antibody according to any one of the preceding claims.

~~98~~. A host cell comprising the nucleic acid of claim ~~87~~.

~~109~~. A method of producing an antibody comprising culturing the host cell of claim 8 so that the antibody is produced.

10 ~~110~~. The method of claim ~~109~~, further comprising recovering the antibody from the host cell.

~~1211~~. A pharmaceutical formulation comprising the antibody according any one of claims 1 to ~~76~~ and a pharmaceutically acceptable carrier.

~~1312~~. The antibody according any one of claims 1 to ~~76~~ for use as a medicament.

~~1413~~. The antibody according any one of claims 1 to ~~76~~ for use in treating cancer.

15 ~~1514~~. Use of the antibody according any one of claims 1 to ~~76~~ in the manufacture of a medicament for treating cancer.