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(54) Title: ANTI-TIM-3 ANTIBODIES

(57) Abstract: Anti-TIM-3 antibodies are disclosed, as well as pharmaceutical compositions comprising such antibodies, and uses and methods using the same, such as in the treatment of cancer, infectious diseases, or T-cell dysfunctional disorders. Bispecific antibodies against TIM-3 and other targets are also disclosed, with preferred embodiment of a bispecific antibody against TIM-3 and CD3.

A3 clone

DIQMTQSPSFMSASVGDRVTITCRASQDIGSYLAWYQQKPGKAPKLLIYAASTL
QSGVPSRFSGSGSGTDFTLTINSLQPEDFATYYCQQSYSSPPTFGPGTTLEIK
(SEQ ID NO:1)

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

B10 clone

EIVLTQSPATLSFSPGERATLSCRASQSVGSYLAWYQQRPGQAPRPLIYDATN
RATGIPTRFSGSGSGTDFTLTISSLEPEDFATYYCQHRRTFGRGTKLEIK
(SEQ ID NO:2)

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

G6 clone

DVVMTQSPLSLPVTPGEPASISCRSSQSLLHSNGYNYLDWYLQKPGQSPQLLI
YLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQGTHWPPTFGQ
GTKVELK (SEQ ID NO:3)

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

Figure 1

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Anti-TIM-3 Antibodies

Field of the Invention

The present invention relates to antibodies that bind to T cell immunoglobulin mucin 3 (TIM-3).

Background to the Invention

T-cell exhaustion is a state of T-cell dysfunction that arises during many chronic infections and cancer. It is defined by poor T-cell effector function, sustained expression of inhibitory receptors and a transcriptional state distinct from that of functional effector or memory T-cells. Exhaustion prevents optimal control of infection and tumors. (E John Wherry., *Nature Immunology* 12, 492-499 (2011)).

T-cell exhaustion is characterized by the stepwise and progressive loss of T-cell functions. Exhaustion is well-defined during chronic lymphocytic choriomeningitis virus infection and commonly develops under conditions of antigen-persistence, which occur following many chronic infections including hepatitis B virus, hepatitis C virus and human immunodeficiency virus infections, as well as during tumor metastasis. Exhaustion is not a uniformly disabled setting as a gradation of phenotypic and functional defects can manifest, and these cells are distinct from prototypic effector, memory and also anergic T cells. Exhausted T cells most commonly emerge during high-grade chronic infections, and the levels and duration of antigenic stimulation are critical determinants of the process. (Yi et al., *Immunology* Apr 2010; 129(4):474-481).

Circulating human tumor-specific $CD8^+$ T cells may be cytotoxic and produce cytokines *in vivo*, indicating that self- and tumor-specific human $CD8^+$ T cells can reach functional competence after potent immunotherapy such as vaccination with peptide, incomplete Freund's adjuvant (IFA), and CpG or after adoptive transfer. In contrast to peripheral blood, T-cells from metastasis are functionally deficient, with abnormally low cytokine production and upregulation of the inhibitory receptors PD-1, CTLA-4, and TIM-3. Functional deficiency is reversible, since T-cells isolated from melanoma tissue can restore IFN-γ production after short-term *in vitro* culture. However, it remains to be determined whether this functional impairment involves further molecular pathways, possibly resembling T-cell exhaustion or anergy as defined in animal models. (Baitsch et al., *J Clin Invest.* 2011;121(6):2350-2360).

Programmed cell death 1 (PD-1), also called CD279, is a type I membrane protein encoded in humans by the *PDCD1* gene. It has two ligands, PD-L1 and PD-L2.

The PD-1 pathway is a key immune-inhibitory mediator of T-cell exhaustion. Blockade of this pathway can lead to T-cell activation, expansion, and enhanced effector functions. As such, PD-1 negatively regulates T cell responses. PD-1 has been identified as a marker of exhausted T cells in chronic disease states, and blockade of PD-1:PD-1L interactions has been shown to partially restore T cell function. (Sakuishi et al., *JEM* Vol. 207, September 27, 2010, pp2187-2194).

Nivolumab (BMS-936558) is an anti–PD-1 that was approved for the treatment of melanoma in Japan in July 2014. Other anti-PD-1 antibodies are described in WO 2010/077634, WO 2006/121168.

T cell immunoglobulin mucin 3 (TIM-3) is an immune regulator identified as being upregulated on exhausted $CD8^+$ T cells (Sakuishi et al., *JEM* Vol. 207, September 27, 2010, pp2187-2194). TIM-3 was originally identified as being selectively expressed on IFN-γ–secreting Th1 and Tc1 cells. Interaction of TIM-3 with its ligand, galectin-9, triggers cell death in $TIM\text{-}3^+$ T cells. Anti-TIM-3 antibodies are described in Ngiow et al (Cancer Res. 2011 May 15;71(10):3540-51),and in US8,552,156.

Both TIM-3 and PD-1 can function as negative regulators of T cell responses and combined targeting of the TIM-3 and PD-1 pathways is more effective in controlling tumor growth than targeting either pathway alone. (Sakuishi et al., *JEM* Vol. 207, September 27, 2010, pp2187-2194; and Ngiow et al *Cancer Res.* 2011 May 15;71(10):3540-51).

TIM-3 can also be expressed on the surface of tumor cells, particularly tumor cells of hematopoietic origin, such for example acute myeloid leukemia cells (Kikushige et al., Cell Stem Cell 2010; 3:7(6)708-17), Therefore in some instances TIM-3 could be a tumor-associated antigen that could be targeted by specific antibodies.

Summary of the Invention

The present invention is concerned with antibodies, or antigen binding fragments, that bind to TIM-3. Heavy and light chain polypeptides are also disclosed. The antibodies, antigen binding fragments and polypeptides may be provided in isolated and/or purified

form and may be formulated into compositions suitable for use in research, therapy and diagnosis.

In some embodiments the antibody, or antigen binding fragment, or polypeptide is cytotoxic, e.g. against TIM-3 expressing cells, such as TIM-3 expressing T-cells or tumour cells. In some embodiments the antibody, or antigen binding fragment, or polypeptide is useful in treating cancer owing to its cytotoxic effect. Suitable cancers include leukemia, such as acute myeloid leukemia.

In some embodiments the antibody, or antigen binding fragment, or polypeptide may be effective to restore T-cell function in T-cells, e.g. $CD8^+$ T-cells, exhibiting T-cell exhaustion or T-cell anergy.

Different aspects of the present invention are based on antibodies designated A3, B10, G6, G7, and G9. Further aspects of the present invention are based on antibodies designated A11 and A11_gl.

A3

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the amino acid sequence of the antibody may comprise the amino acid sequences i) to iii), or the amino acid sequences iv) to vi), or preferably the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
ii)	LC-CDR2:	AASTLQS	(SEQ ID NO:7)
iii)	LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)
iv)	HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24) or
		SYYMH	(SEQ ID NO:58)
v)	HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
vi)	HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

In connection with all aspects of the present invention, in embodiments wherein HC-CDR1: SYYMH (SEQ ID NO:58), this sequence may be comprised in the larger sequence GYTFTSYYMH (SEQ ID NO: 24).

The antibody, or antigen binding fragment, may comprise at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

The antibody, or antigen binding fragment, may comprise at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

The antibody may comprise at least one light chain variable region incorporating the CDRs shown in Figures 1 or 3. The antibody may comprise at least one heavy chain variable region incorporating the CDRs shown in Figures 2 or 3.

The antibody may comprise at least one light chain variable region (V_L) comprising the amino acid sequence of one of SEQ ID NOs 1, 6, 7, 8 or one of the amino acid sequences shown in Figure 1 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 1, 6, 7, 8 or to the amino acid sequence of the V_L chain amino acid sequence shown in Figure 1.

The antibody may comprise at least one heavy chain variable region (V_H) comprising the amino acid sequence of one of SEQ ID NOs 19, 24 or 58, 25, 26 or one of the amino acid sequences shown in Figure 2 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 19, 24 or 58, 25, 26 or to the amino acid sequence of the V_H chain amino acid sequence shown in Figure 2.

The antibody may comprise at least one light chain variable region comprising the amino acid sequence of one of SEQ ID NOs 1, 6, 7, 8 or one of the amino acid sequences shown in Figure 1 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%, sequence identity to one of SEQ ID NOs 1, 6, 7, 8 or to one of the amino acid sequences of the V_L chain amino acid sequence shown in Figure 1) and at least one heavy chain variable region comprising the amino acid sequence of one of SEQ ID NOs 19, 24 or 58, 25, 26 or one of the amino acid sequence shown in Figure 2 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 19, 24 or 58, 25, 26 or to one of the amino acid sequences of the V_H chain amino acid sequence shown in Figure 2).

The antibody may optionally bind TIM-3. The antibody may optionally have amino acid sequence components as described above. The antibody may be an IgG. In one embodiment an *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, as described herein, bound to TIM-3 is provided.

In one aspect of the present invention an isolated heavy chain variable region polypeptide is provided, the heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the antibody, or antigen binding fragment, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GYTFTSYYMH (SEQ ID NO:24) or SYYMH (SEQ ID NO:58), IINPSGGSTSYAQKFQG (SEQ ID NO:25), SPGVVTALFDY (SEQ ID NO:26) respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQDIGSYLA (SEQ ID NO:6), AASTLQS (SEQ ID NO:7), QQSYSSPPT (SEQ ID NO:8), respectively.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In another aspect of the present invention an antibody, or antigen binding fragment, optionally isolated, is provided comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 19, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:1.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region heavy chain framework sequences between the CDRs according to the arrangement HCFR1:HC-CDR1:HCFR2:HC-CDR2:HCFR3:HC-CDR3:HCFR4. The framework sequences may be derived from human consensus framework sequences.

In one aspect of the present invention an isolated light chain variable region polypeptide, optionally in combination with a heavy chain variable region polypeptide as described herein, is provided, the light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region light chain framework sequences between the CDRs according to the arrangement LCFR1:LC-CDR1:LCFR2:LC-CDR2:LCFR3:LC-CDR3:LCFR4. The framework sequences may be derived from human consensus framework sequences.

<u>B10</u>

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the amino acid sequence of the antibody may comprise the amino acid

sequences i) to iii), or the amino acid sequences iv) to vi), or preferably the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
ii)	LC-CDR2:	DATNRAT	(SEQ ID NO:10)
iii)	LC-CDR3:	QHRRT	(SEQ ID NO:11)
iv)	HC-CDR1:	GGSIGSSDYYWG	(SEQ ID NO:27), or
		SSDYYWG	(SEQ ID NO:59)
v)	HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
vi)	HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

In connection with all aspects of the present invention, in embodiments wherein HC-CDR1: SSDYYWG (SEQ ID NO:59), this sequence may be comprised in the larger sequence GGSIGSSDYYWG (SEQ ID NO:27).

The antibody, or antigen binding fragment, may comprise at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

The antibody, or antigen binding fragment, may comprise at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GGSIGSSDYYWG	(SEQ ID NO:27), or
	SSDYYWG	(SEQ ID NO:59)
HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

The antibody may comprise at least one light chain variable region incorporating the CDRs shown in Figures 1 or 3. The antibody may comprise at least one heavy chain variable region incorporating the CDRs shown in Figures 2 or 3.

The antibody may comprise at least one light chain variable region (V_L) comprising the amino acid sequence of one of SEQ ID NOs 2, 9, 10, 11 or one of the amino acid sequences shown in Figure 1 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 2, 9, 10, 11 or to the amino acid sequence of the V_L chain amino acid sequence shown in Figure 1.

The antibody may comprise at least one heavy chain variable region (V_H) comprising the amino acid sequence of one of SEQ ID NOs 20, 27 or 59, 28, 29 or one of the amino acid sequences shown in Figure 2 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 20, 27 or 59, 28, 29 or to the amino acid sequence of the V_H chain amino acid sequence shown in Figure 2.

The antibody may comprise at least one light chain variable region comprising the amino acid sequence of one of SEQ ID NOs 2, 9, 10, 11 or one of the amino acid sequences shown in Figure 1 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%, sequence identity to one of SEQ ID NOs 2, 9, 10, 11 or to one of the amino acid sequences of the V_L chain amino acid sequence shown in Figure 1) and at least one heavy chain variable region comprising the amino acid sequence of one of SEQ ID NOs 20, 27 or 59, 28, 29 or one of the amino acid sequence shown in Figure 2 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 20, 27 or 59, 28, 29 or to one of the amino acid sequences of the V_H chain amino acid sequence shown in Figure 2).

The antibody may optionally bind TIM-3. The antibody may optionally have amino acid sequence components as described above. The antibody may be an IgG. In one embodiment an *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, as described herein, bound to TIM-3 is provided.

In one aspect of the present invention an isolated heavy chain variable region polypeptide is provided, the heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GGSIGSSDYYWG	(SEQ ID NO:27) , or
	SSDYYWG	(SEQ ID NO:59)
HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the antibody, or antigen binding fragment, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GGSIGSSDYYWG (SEQ ID NO:27) or SSDYYWG (SEQ ID NO:59), SIYYSGSTYYNPSLKS (SEQ ID NO:28), GEHRGEFDY (SEQ ID NO:29), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQSVGSYLA (SEQ ID NO:9), DATNRAT (SEQ ID NO:10), QHRRT (SEQ ID NO:11), respectively.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In another aspect of the present invention an antibody, or antigen binding fragment, optionally isolated, is provided comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 20, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:2.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region heavy chain framework sequences between the CDRs according to the arrangement HCFR1:HC-CDR1:HCFR2:HC-CDR2:HCFR3:HC-CDR3:HCFR4. The framework sequences may be derived from human consensus framework sequences.

In one aspect of the present invention an isolated light chain variable region polypeptide, optionally in combination with a heavy chain variable region polypeptide as described

herein, is provided, the light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region light chain framework sequences between the CDRs according to the arrangement LCFR1:LC-CDR1:LCFR2:LC-CDR2:LCFR3:LC-CDR3:LCFR4. The framework sequences may be derived from human consensus framework sequences.

G6

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the amino acid sequence of the antibody may comprise the amino acid sequences i) to iii), or the amino acid sequences iv) to vi), or preferably the amino acid sequences i) to vi):

i)	LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
ii)	LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
iii)	LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)
iv)	HC-CDR1:	GGSISSSNWWS	(SEQ ID NO:30), or
		SSNWWS	(SEQ ID NO:60)
v)	HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
vi)	HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

In connection with all aspects of the present invention, in embodiments wherein HC-CDR1: SSNWWS (SEQ ID NO:60), this sequence may be comprised in the larger sequence GGSISSSNWWS (SEQ ID NO: 30).

The antibody, or antigen binding fragment, may comprise at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)

LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

The antibody, or antigen binding fragment, may comprise at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GGSISSSNWWS	(SEQ ID NO:30), or
	SSNWWS	(SEQ ID NO:60)
HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

The antibody may comprise at least one light chain variable region incorporating the CDRs shown in Figures 1 or 3. The antibody may comprise at least one heavy chain variable region incorporating the CDRs shown in Figures 2 or 3.

The antibody may comprise at least one light chain variable region (V_L) comprising the amino acid sequence of one of SEQ ID NOs 3, 12, 13, 14 or one of the amino acid sequences shown in Figure 1 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 3, 12, 13, 14 or to the amino acid sequence of the V_L chain amino acid sequence shown in Figure 1.

The antibody may comprise at least one heavy chain variable region (V_H) comprising the amino acid sequence of one of SEQ ID NOs 21, 30 or 60, 31, 32 or one of the amino acid sequences shown in Figure 2 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 21, 30 or 60, 31, 32 or to the amino acid sequence of the V_H chain amino acid sequence shown in Figure 2.

The antibody may comprise at least one light chain variable region comprising the amino acid sequence of one of SEQ ID NOs 3, 12, 13, 14 or one of the amino acid sequences shown in Figure 1 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%, sequence identity to one of SEQ ID NOs 3, 12, 13, 14or to one of the amino acid sequences of the V_L chain amino acid sequence shown in Figure 1) and at least one heavy chain variable region

comprising the amino acid sequence of one of SEQ ID NOs 21, 30 or 60, 31, 32 or one of the amino acid sequence shown in Figure 2 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 21, 30 or 60, 31, 32 or to one of the amino acid sequences of the V_H chain amino acid sequence shown in Figure 2).

The antibody may optionally bind TIM-3. The antibody may optionally have amino acid sequence components as described above. The antibody may be an IgG. In one embodiment an *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, as described herein, bound to TIM-3 is provided.

In one aspect of the present invention an isolated heavy chain variable region polypeptide is provided, the heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GGSISSSNWWS	(SEQ ID NO:30), or
	SSNWWS	(SEQ ID NO:60)
HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the antibody, or antigen binding fragment, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GGSISSSNWWS (SEQ ID NO:30) or SSNWWS (SEQ ID NO:60), EIYHSGSTNYNPSLKS (SEQ ID NO:31), VVAVAGTVDY (SEQ ID NO:32), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RSSQSLLHSNGYNYLD (SEQ ID NO:12), LGSNRAS (SEQ ID NO:13), MQGTHWPPT (SEQ ID NO:14),respectively.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In another aspect of the present invention an antibody, or antigen binding fragment, optionally isolated, is provided comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain

sequence: SEQ ID NO: 21, and
the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:3.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region heavy chain framework sequences between the CDRs according to the arrangement HCFR1:HC-CDR1:HCFR2:HC-CDR2:HCFR3:HC-CDR3:HCFR4. The framework sequences may be derived from human consensus framework sequences.

In one aspect of the present invention an isolated light chain variable region polypeptide, optionally in combination with a heavy chain variable region polypeptide as described herein, is provided, the light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region light chain framework sequences between the CDRs according to the arrangement LCFR1:LC-CDR1:LCFR2:LC-CDR2:LCFR3:LC-CDR3:LCFR4. The framework sequences may be derived from human consensus framework sequences.

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In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the amino acid sequence of the antibody may comprise the amino acid sequences i) to iii), or the amino acid sequences iv) to vi), or preferably the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
ii)	LC-CDR2:	GASSRAT	(SEQ ID NO:16)
iii)	LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)
iv)	HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
		SYYMH	(SEQ ID NO:58)

v) HC-CDR2: IINPSGGSTSYAQKFQG (SEQ ID NO:25)

vi) HC-CDR3: DQYSSGWYYYGMDV (SEQ ID NO:33)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

In connection with all aspects of the present invention, in embodiments wherein HC-CDR1: SYYMH (SEQ ID NO:58), this sequence may be comprised in the larger sequence GYTFTSYYMH (SEQ ID NO: 24).

The antibody, or antigen binding fragment, may comprise at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

The antibody, or antigen binding fragment, may comprise at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

The antibody may comprise at least one light chain variable region incorporating the CDRs shown in Figures 1 or 3. The antibody may comprise at least one heavy chain variable region incorporating the CDRs shown in Figures 2 or 3.

The antibody may comprise at least one light chain variable region (V_L) comprising the amino acid sequence of one of SEQ ID NOs 4, 15, 16, 17 or one of the amino acid sequences shown in Figure 1 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 4, 15, 16, 17 or to the amino acid sequence of the V_L chain amino acid sequence shown in Figure 1.

The antibody may comprise at least one heavy chain variable region (V_H) comprising the amino acid sequence of one of SEQ ID NOs 22, 24 or 58, 25, 33 or one of the amino acid sequences shown in Figure 2 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 22, 24 or 58, 25, 33 or to the amino acid sequence of the V_H chain amino acid sequence shown in Figure 2.

The antibody may comprise at least one light chain variable region comprising the amino acid sequence of one of SEQ ID NOs 4, 15, 16, 17 or one of the amino acid sequences shown in Figure 1 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%, sequence identity to one of SEQ ID NOs 4, 15, 16, 17 or to one of the amino acid sequences of the V_L chain amino acid sequence shown in Figure 1) and at least one heavy chain variable region comprising the amino acid sequence of one of SEQ ID NOs 22, 24 or 58, 25, 33 or one of the amino acid sequence shown in Figure 2 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 22, 24 or 58, 25, 33 or to one of the amino acid sequences of the V_H chain amino acid sequence shown in Figure 2).

The antibody may optionally bind TIM-3. The antibody may optionally have amino acid sequence components as described above. The antibody may be an IgG. In one embodiment an *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, as described herein, bound to TIM-3 is provided.

In one aspect of the present invention an isolated heavy chain variable region polypeptide is provided, the heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24) , or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the antibody, or antigen binding fragment, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GYTFTSYYMH (SEQ ID NO:24) or SYYMH (SEQ ID NO:58), IINPSGGSTSYAQKFQG (SEQ ID NO:25), DQYSSGWYYYGMDV (SEQ ID NO:33), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQSVSSSYLA (SEQ ID NO:15), GASSRAT (SEQ ID NO:16), QQYGSSPIT (SEQ ID NO:17), respectively.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In another aspect of the present invention an antibody, or antigen binding fragment, optionally isolated, is provided comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 22, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:4.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region heavy chain framework sequences between the CDRs according to the arrangement HCFR1:HC-CDR1:HCFR2:HC-CDR2:HCFR3:HC-CDR3:HCFR4. The framework sequences may be derived from human consensus framework sequences.

In one aspect of the present invention an isolated light chain variable region polypeptide, optionally in combination with a heavy chain variable region polypeptide as described herein, is provided, the light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region light chain framework sequences between the CDRs according to the arrangement LCFR1:LC-CDR1:LCFR2:LC-CDR2:LCFR3:LC-CDR3:LCFR4. The framework sequences may be derived from human consensus framework sequences.

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In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the amino acid sequence of the antibody may comprise the amino acid sequences i) to iii), or the amino acid sequences iv) to vi), or preferably the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
ii)	LC-CDR2:	GASSRAT	(SEQ ID NO:16)
iii)	LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)
iv)	HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
		SYYMH	(SEQ ID NO:58)
v)	HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
vi)	HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

In connection with all aspects of the present invention, in embodiments wherein HC-CDR1: SYYMH (SEQ ID NO:58), this sequence may be comprised in the larger sequence GYTFTSYYMH (SEQ ID NO: 24).

The antibody, or antigen binding fragment, may comprise at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

The antibody, or antigen binding fragment, may comprise at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

The antibody may comprise at least one light chain variable region incorporating the CDRs shown in Figures 1 or 3. The antibody may comprise at least one heavy chain variable region incorporating the CDRs shown in Figures 2 or 3.

The antibody may comprise at least one light chain variable region (V_L) comprising the amino acid sequence of one of SEQ ID NOs 5, 15, 16, 17 or one of the amino acid sequences shown in Figure 1 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 5, 15, 16, 17 or to the amino acid sequence of the V_L chain amino acid sequence shown in Figure 1.

The antibody may comprise at least one heavy chain variable region (V_H) comprising the amino acid sequence of one of SEQ ID NOs 23, 24 or 58, 25, 34 or one of the amino acid sequences shown in Figure 2 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 23, 24 or 58, 25, 34 or to the amino acid sequence of the V_H chain amino acid sequence shown in Figure 2.

The antibody may comprise at least one light chain variable region comprising the amino acid sequence of one of SEQ ID NOs 5, 15, 16, 17 or one of the amino acid sequences shown in Figure 1 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%, sequence identity to one of SEQ ID NOs 5, 15, 16, 17 or to one of the amino acid sequences of the V_L chain amino acid sequence shown in Figure 1) and at least one heavy chain variable region comprising the amino acid sequence of one of SEQ ID NOs 23, 24 or 58, 25, 34 or one of the amino acid sequence shown in Figure 2 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of

SEQ ID NOs 23, 24 or 58, 25, 34 or to one of the amino acid sequences of the V_H chain amino acid sequence shown in Figure 2).

The antibody may optionally bind TIM-3. The antibody may optionally have amino acid sequence components as described above. The antibody may be an IgG. In one embodiment an *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, as described herein, bound to TIM-3 is provided.

In one aspect of the present invention an isolated heavy chain variable region polypeptide is provided, the heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the antibody, or antigen binding fragment, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GYTFTSYYMH (SEQ ID NO:24) or SYYMH (SEQ ID NO:58), IINPSGGSTSYAQKFQG (SEQ ID NO:25), DLYSYGFYYYGMDV (SEQ ID NO:34), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQSVSSSYLA (SEQ ID NO:15), GASSRAT (SEQ ID NO:16), QQYGSSPIT (SEQ ID NO:17), respectively.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In another aspect of the present invention an antibody, or antigen binding fragment, optionally isolated, is provided comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 23, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:5.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region heavy chain framework sequences between the CDRs according to the arrangement HCFR1:HC-CDR1:HCFR2:HC-CDR2:HCFR3:HC-CDR3:HCFR4. The framework sequences may be derived from human consensus framework sequences.

In one aspect of the present invention an isolated light chain variable region polypeptide, optionally in combination with a heavy chain variable region polypeptide as described herein, is provided, the light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region light chain framework sequences between the CDRs according to the arrangement LCFR1:LC-CDR1:LCFR2:LC-CDR2:LCFR3:LC-CDR3:LCFR4. The framework sequences may be derived from human consensus framework sequences.

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In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the amino acid sequence of the antibody may comprise the amino acid sequences i) to iii), or the amino acid sequences iv) to vi), or preferably the amino acid sequences i) to vi):

i)	LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
ii)	LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
iii)	LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)
iv)	HC-CDR1:	GGSFSGYYWS	(SEQ ID NO:52), or
		GYYWS	(SEQ ID NO:61)
v)	HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)

vi) HC-CDR3: GYVAGFDY (SEQ ID NO:54)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

In connection with all aspects of the present invention, in embodiments wherein HC-CDR1: GYYWS (SEQ ID NO:61), this sequence may be comprised in the larger sequence GGSFSGYYWS (SEQ ID NO:52).

The antibody, or antigen binding fragment, may comprise at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

The antibody, or antigen binding fragment, may comprise at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GGSFSGYYWS	(SEQ ID NO:52), or
	GYYWS	(SEQ ID NO:61)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

The antibody may comprise at least one light chain variable region incorporating the CDRs shown in Figures 1 or 3. The antibody may comprise at least one heavy chain variable region incorporating the CDRs shown in Figures 2 or 3.

The antibody may comprise at least one light chain variable region (V_L) comprising the amino acid sequence of one of SEQ ID NOs 45, 46, 47, 48, 49 or one of the amino acid sequences shown in Figure 1 or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 45, 46, 47, 48, 49 or to one of the amino acid sequences of the V_L chain amino acid sequences shown in Figure 1.

The antibody may comprise at least one heavy chain variable region (V_H) comprising the amino acid sequence of one of SEQ ID NOs 50, 51, 52 or 61, 53, 54 or one of the amino acid sequences shown in Figure 2 or an amino acid sequence having at least 70%, more

preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 50, 51, 52 or 61, 53, 54 or to one of the amino acid sequences of the V_H chain amino acid sequences shown in Figure 2.

The antibody may comprise at least one light chain variable region comprising the amino acid sequence of one of SEQ ID NOs 45, 46, 47, 48, 49 or one of the amino acid sequences shown in Figure 1 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 90%, 95%, 96%, 97%, 98%, 99% or 100%, sequence identity to one of SEQ ID NOs 45, 46, 47, 48, 49 or to one of the amino acid sequences of the V_L chain amino acid sequences shown in Figure 1) and at least one heavy chain variable region comprising the amino acid sequence of one of SEQ ID NOs 50, 51, 52 or 61, 53, 54 or one of the amino acid sequence shown in Figure 2 (or an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to one of SEQ ID NOs 50, 51, 52 or 61, 53, 54 or to one of the amino acid sequences of the V_H chain amino acid sequences shown in Figure 2).

The antibody may optionally bind TIM-3. The antibody may optionally have amino acid sequence components as described above. The antibody may be an IgG. In one embodiment an *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, as described herein, bound to TIM-3 is provided.

In one aspect of the present invention an isolated heavy chain variable region polypeptide is provided, the heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GGSFSGYYWS	(SEQ ID NO:52), or
	GYYWS	(SEQ ID NO:61)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

In one aspect of the present invention an antibody, or antigen binding fragment, is provided, the antibody, or antigen binding fragment, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GGSFSGYYWS (SEQ ID NO:52) or GYYWS

(SEQ ID NO:61), EINHSGSTNYNPSLKS (SEQ ID NO:53), GYVAGFDY (SEQ ID NO:54), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3,, having at least 85% overall sequence identity to SGSSSNIGNNYVS (SEQ ID NO:47), GNNWRPS (SEQ ID NO:48), ETWDSSLSAGV (SEQ ID NO:49), respectively.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In another aspect of the present invention an antibody, or antigen binding fragment, optionally isolated, is provided comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO:50 or 51, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:45 or 46.

In some embodiments the degree of sequence identity may be one of 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region heavy chain framework sequences between the CDRs according to the arrangement HCFR1:HC-CDR1:HCFR2:HC-CDR2:HCFR3:HC-CDR3:HCFR4. The framework sequences may be derived from human consensus framework sequences.

In one aspect of the present invention an isolated light chain variable region polypeptide, optionally in combination with a heavy chain variable region polypeptide as described herein, is provided, the light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

In some embodiments the antibody, antigen binding fragment, or polypeptide further comprises variable region light chain framework sequences between the CDRs according to the arrangement LCFR1:LC-CDR1:LCFR2:LC-CDR2:LCFR3:LC-CDR3:LCFR4. The framework sequences may be derived from human consensus framework sequences.

In some embodiments, the antibody, or antibody binding fragment, may further comprise a human constant region. For example selected from one of IgG1, IgG2, IgG3 and IgG4.

In some embodiments, the antibody, or antibody binding fragment, may further comprise a murine constant region. For example, selected from one of IgG1, IgG2A, IgG2B and IgG3.

In another aspect of the present invention, an antibody or antigen binding fragment is provided, optionally isolated, which is capable of binding to TIM-3, and which is a bispecific antibody or a bispecific antigen binding fragment. In some embodiments, the bispecific antibody or bispecific antigen binding fragment comprises an antigen binding fragment or polypeptide capable of binding to TIM-3 as described herein, and additionally comprises an antigen binding domain which is capable of binding to another target protein, e.g. a target protein other than TIM-3. In some embodiments, the target protein is a cell surface receptor. In some embodiments, the target protein is a cell surface receptor expressed on the cell surface of immune cells, e.g. T cells. In some embodiments, the antigen binding domain capable of binding to another target protein may be capable of binding to a T cell receptor (TCR) complex or a component thereof. In some embodiments, the antigen binding domain may be capable of binding to CD3 or a CD3 polypeptide. In some embodiments, the antigen binding domain may be capable of binding to one or more of the CD3 polypeptides CD3γ, CD3δ, CD3ζ, or CD3ε. In some embodiments the bispecific antibody is a bispecific T-cell engager antibody. In some embodiments, the target protein may be a member of the CD28 family. In some embodiments, the member of the CD28 family is selected from PD-1, LAG3, ICOS, CTLA4, BTLA or CD28.

In another aspect of the present invention, a composition, e.g. a pharmaceutical composition or medicament, is provided. The composition may comprise an antibody, antigen binding fragment, or polypeptide as described herein and at least one pharmaceutically-acceptable carrier, excipient, adjuvant or diluent.

In another aspect of the present invention an isolated nucleic acid encoding an antibody, antigen binding fragment, or polypeptide as described herein is provided. The nucleic

acid may have a sequence of one of SEQ ID NOs 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 55, 56 or 57 (Figure 4) or a coding sequence which is degenerate as a result of the genetic code, or may have a nucleotide sequence having at least 70% identity thereto, optionally one of 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%.

In one aspect of the present invention there is provided a vector comprising a nucleic acid described herein. In another aspect of the present invention, there is provided a host cell comprising the vector. For example, the host cell may be eukaryotic, or mammalian, e.g. Chinese Hamster Ovary (CHO), or human or may be a prokaryotic cell, e.g. *E. coli.*
In one aspect of the present invention a method for making an antibody, or antigen binding fragment or polypeptide as described herein is provided, the method comprising culturing a host cell as described herein under conditions suitable for the expression of a vector encoding the antibody, or antigen binding fragment or polypeptide, and recovering the antibody, or antigen binding fragment or polypeptide.

In another aspect of the present invention an antibody, antigen binding fragment or polypeptide is provided for use in therapy, or in a method of medical treatment. In another aspect of the present invention an antibody, antigen binding fragment or polypeptide as described herein is provided for use in the treatment of cancer or a T-cell dysfunctional disorder. In another aspect of the present invention, the use of an antibody, antigen binding fragment or polypeptide as described herein in the manufacture of a medicament or pharmaceutical composition for use in the treatment of cancer or a T-cell dysfunctional disorder is provided.

In another aspect a method, *in vitro* or *in vivo*, of killing a cell that expresses TIM-3 is provided, the method comprising administering an antibody, antigen binding fragment or polypeptide as described herein to a cell that expresses (or overexpresses) TIM-3. The cell may be a cancer cell, e.g. leukemia or acute myeloid leukemia cell, white blood cell or T-cell. In some embodiments, the acute myeloid leukemia cell may be a stem cell; for example, in some embodiments the acute myeloid leukemia cell may be CD34+.

In another aspect of the present invention a method of enhancing T-cell function comprising administering an antibody, antigen binding fragment or polypeptide as described herein to a dysfunctional T-cell is provided. The method may be performed *in vitro* or *in vivo*.

In another aspect of the present invention a method of treating cancer or a T-cell dysfunctional disorder or an infectious disease is provided, the method comprising administering an antibody, antigen binding fragment or polypeptide as described herein to a patient suffering from cancer or a T-cell dysfunctional disorder.

In another aspect of the present invention a method of treating an infectious disease is provided, the method comprising administering an antibody, antigen binding fragment of polypeptide as described herein to a patient suffering from an infectious disease.

In another aspect of the present invention a method of modulating an immune response in a subject is provided, the method comprising administering to the subject an antibody, antigen binding fragment or polypeptide as described herein such that the immune response in the subject is modulated.

In another aspect of the present invention a method of inhibiting growth of tumor cells in a subject is provided, the method comprising administering to the subject a therapeutically effective amount of an antibody, antigen binding fragment or polypeptide as described herein.

In another aspect of the present invention a method is provided, the method comprising contacting a sample containing, or suspected to contain, TIM-3 with an antibody or antigen binding fragment, as described herein, and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.

In another aspect of the present invention a method of diagnosing a disease or condition in a subject is provided, the method comprising contacting, *in vitro*, a sample from the subject with an antibody, or antigen binding fragment, , as described herein, and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.
An aspect of the present invention is a method of selecting a patient for treatment with a modulator of TIM3 signalling, such as an anti-TIM3 antibody or anti-TIM3 agent, the method comprising contacting, *in vitro*, a sample from the subject with an antibody, or antigen binding fragment as described herein, and detecting the formation of a complex of the antibody, or antigen binding fragment, and TIM-3.

In a further aspect of the present invention a method of selecting or stratifying a subject for treatment with a modulator of TIM-3 signalling is provided, the method comprising contacting, *in vitro*, a sample from the subject with an antibody, or antigen binding fragment, according to the present invention and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.

In a further aspect of the present invention the use of an antibody, or antigen binding fragment, as described herein, for the detection of TIM-3 *in vitro* is provided. In another aspect of the present invention the use of an antibody, or antigen binding fragment, as described herein, as an *in vitro* diagnostic agent is provided.

In a further aspect of the present invention a method for expanding a population of T cells is provided, wherein T cells are contacted *in vitro* or *ex vivo* with an antibody, antigen binding fragment or polypeptide according to the present invention.

In a further aspect of the present invention a method of treatment of a subject having a T-cell dysfunctional disorder is provided, the method comprising culturing T cells obtained from a blood sample from a subject in the presence of an antibody, antigen binding fragment or polypeptide according to the present invention so as to expand the T cell population, collecting expanded T cells, and administering the expanded T cells to a subject in need of treatment.

In methods of the present invention the antibody, antigen binding fragment or polypeptide may be provided as a composition as described herein.

In some embodiments the antibody may be one of clones A3, B10, G6, G7, G9, A11 or A11_gl.

Description

Antibodies

Antibodies according to the present invention preferably bind to TIM-3 (the antigen), preferably human or rhesus TIM-3, optionally with a K_D in the range 0.1 to 2nM.

In any aspect of the present invention the antibody preferably specifically binds TIM-3 (e.g. human or rhesus).

Antibodies according to the present invention may be provided in isolated form.

Antibodies according to the present invention may exhibit least one of the following properties:

a) binds to human TIM-3 with a K_D of 1μM or less, preferably one of ≤ 1μM, ≤ 100nM, ≤10nM, ≤1nM or ≤100pM;

b) is cytotoxic against TIM-3 expressing cells (antibody dependent cell-mediated cytotoxicity, ADCC), e.g. TIM-3 expressing acute myeloid leukemia cells

c) increases T -cell proliferation in a Mixed Lymphocyte Reaction (MLR) assay (e.g. see Bromelow et al *J.Immunol Methods*, 2001 Jan 1;247(1-2):1-8);

d) increases interferon-gamma production in an MLR assay; or

e) increases interleukin-2 (IL-2) secretion in an MLR assay.

By “antibody” we include a fragment or derivative thereof, or a synthetic antibody or synthetic antibody fragment.

In view of today's techniques in relation to monoclonal antibody technology, antibodies can be prepared to most antigens. The antigen-binding portion may be a part of an antibody (for example a Fab fragment) or a synthetic antibody fragment (for example a single chain Fv fragment [ScFv]). Suitable monoclonal antibodies to selected antigens may be prepared by known techniques, for example those disclosed in "Monoclonal Antibodies: A manual of techniques ", H Zola (CRC Press, 1988) and in "Monoclonal Hybridoma Antibodies: Techniques and Applications ", J G R Hurrell (CRC Press, 1982). Chimaeric antibodies are discussed by Neuberger et al (1988, 8th International Biotechnology Symposium Part 2, 792-799).

Monoclonal antibodies (mAbs) are useful in the methods of the invention and are a homogenous population of antibodies specifically targeting a single epitope on an antigen.

Polyclonal antibodies are useful in the methods of the invention. Monospecific polyclonal antibodies are preferred. Suitable polyclonal antibodies can be prepared using methods well known in the art.

Antigen binding fragments of antibodies, such as Fab and Fab_2 fragments may also be

used/provided as can genetically engineered antibodies and antibody fragments. The variable heavy (V_H) and variable light (V_L) domains of the antibody are involved in antigen recognition, a fact first recognised by early protease digestion experiments. Further confirmation was found by "humanisation" of rodent antibodies. Variable domains of rodent origin may be fused to constant domains of human origin such that the resultant antibody retains the antigenic specificity of the rodent parented antibody (Morrison et al (1984) Proc. Natl. Acad. Sd. USA 81, 6851-6855).

That antigenic specificity is conferred by variable domains and is independent of the constant domains is known from experiments involving the bacterial expression of antibody fragments, all containing one or more variable domains. These molecules include Fab-like molecules (Better et al (1988) Science 240, 1041); Fv molecules (Skerra et al (1988) Science 240, 1038); single-chain Fv (ScFv) molecules where the V_H and V_L partner domains are linked via a flexible oligopeptide (Bird et al (1988) Science 242, 423; Huston et al (1988) Proc. Natl. Acad. Sd. USA 85, 5879) and single domain antibodies (dAbs) comprising isolated V domains (Ward et al (1989) Nature 341, 544). A general review of the techniques involved in the synthesis of antibody fragments which retain their specific binding sites is to be found in Winter & Milstein (1991) Nature 349, 293- 299.

By "ScFv molecules" we mean molecules wherein the V_H and V_L partner domains are covalently linked, e.g. by a flexible oligopeptide.

Fab, Fv, ScFv and dAb antibody fragments can all be expressed in and secreted from E. coli, thus allowing the facile production of large amounts of the said fragments.

Whole antibodies, and $F(ab')_2$ fragments are "bivalent". By "bivalent" we mean that the said antibodies and $F(ab')_2$ fragments have two antigen combining sites. In contrast, Fab, Fv, ScFv and dAb fragments are monovalent, having only one antigen combining site. Synthetic antibodies which bind to TIM-3 may also be made using phage display technology as is well known in the art.

Aspects of the present invention include bi-specific antibodies, e.g. composed of two different fragments of two different antibodies, such that the bi-specific antibody binds two types of antigen. One of the antigens is TIM-3, the bi-specific antibody comprising a fragment as described herein that binds to TIM-3. The antibody may contain a different fragment having affinity for a second antigen, which may be any desired antigen, for

example CD3 which has been used in cancer immunotherapy to bind to cytotoxic cells, recruit and target them to the site of a tumor. Techniques for the preparation of bi-specific antibodies are well known in the art, e.g. see Mueller, D et al., (2010 *Biodrugs* **24** (2): 89–98), Wozniak-Knopp G et al., (2010 *Protein Eng Des* **23** (4): 289–297. Baeuerle, PA et al., (2009 *Cancer Res* **69** (12): 4941–4944).

Accordingly, the present invention provides an antibody or antigen binding fragment which is capable of binding to TIM-3, and which is a bispecific antibody or a bispecific antigen binding fragment. In some embodiments, the bispecific antibody or bispecific antigen binding fragment may be isolated.

In some embodiments, the bispecific antibodies and bispecific antigen binding fragments comprise an antigen binding fragment or a polypeptide according to the present invention. In some embodiments, the bispecific antibodies and bispecific antigen binding fragments comprise an antigen binding domain capable of binding to TIM-3, wherein the antigen binding domain which is capable of binding to TIM-3 comprises or consists of an antigen binding fragment or a polypeptide according to the present invention.

In some embodiments the bispecific antibodies and bispecific antigen binding fragments comprise an antigen binding domain capable of binding to TIM-3, and an antigen binding domain capable of binding to another target protein.

The antigen binding domain capable of binding to another target protein may be capable of binding to another protein other than TIM-3. In some embodiments, the target protein is a cell surface receptor. In some embodiments, the target protein is a cell surface receptor expressed on the cell surface of immune cells. In some embodiments, the target protein is a cell surface receptor expressed on the cell surface of T cells.

In some embodiments, the antigen binding domain capable of binding to another target protein may be capable of binding to a T cell receptor (TCR) complex or a component thereof. In some embodiments, the antigen binding domain may be capable of binding to CD3 or a CD3 polypeptide. In some embodiments, the antigen binding domain may be capable of binding to one or more of the CD3 polypeptides CD3γ, CD3δ, CD3ζ, or CD3ε.In some embodiments the bispecific antibody is a bispecific T-cell engager antibody.

In some embodiments the bispecific antibody or fragment is capable of directing T cell activity (e.g. cytotoxic activity) against a TIM-3 expressing cell. That is, in some embodiments, T cell activity (e.g. cytotoxic activity) against a TIM-3 expressing cell is increased in the presence of the bispecific antibody or fragment (e.g. relative to activity against a TIM-3 expressing cell in the absence of the bispecific antibody or fragment). T cell activity against a TIM-3 expressing cell can be determined *in vitro* by methods well known to the skilled person, for example by incubating T cells with TIM-3 expressing cells and measuring cell lysis as described herein.

In some embodiments, the bispecific antibody is provided as a fusion protein of two single-chain variable fragments (scFV) format, comprising a V_H and V_L of a TIM-3 binding antibody or antibody fragment according to the present invention, and a V_H and V_L of an antibody or antibody fragment capable of binding to CD3 or a CD3 polypeptide.

In some embodiments, the antigen binding domain for CD3 or a CD3 polypeptide may comprise the CDRs, light and heavy chain variable domains or other CD3- or CD3 polypeptide- binding fragment of e.g. anti-CD3 antibody clone OKT3 (eBioscience), clone CD3-12 (AbD Serotec), clone UCHT1 (Southern Biotech) clone SP7 (Thermo Scientific Pierce Antibodies), clone SPV-T3b (Thermo Fisher Scientific), clone S4.1 (7D6) (Thermo Fisher Scientific), clone MEM-57 (AbD Serotec), clone 37895 (Miltenyi Biotec), clone CA-3 (Abcam), clone 4D10A6 (Abbiotec), clone HIT3a (Abbiotec), clone LT3 (Source BioScience), clone B-B11 (MyBioSource.com), clone 17A2 (Novus Biologicals), clone BC3 (BioLegend), clone HAM25-1352(MBL International), clone CA-3 (Bosterbio), clone RBT-CD3 (Lifespan BioSciences), Ham25-1157 (Merck Millipore), clone CRIS-7 (Peninsula Laboratories International), clone 5B2, clone 2Q1160 (Santa Cruz Biotechnology), clone M01, clone B1.1 (Abnova Corporation), clone EP449E (BioGenex), clone 6B8D1G5, clone 6B1C12F3 (Sino Biological), clone CL1297 (Atlas Antibodies), clone CC23 (Creative Diagnostics), clone TR66 (Enzo Life Sciences), clone MEM-92 (Cedarlane), clone EPR4516 (Origene Technologies), clone 3A12H2 (Proteintech Group), clone 33-2A3 (ALPCO), clone E272 (Biocare Medical), clone SP162, clone MRQ-39 (Sigma Aldrich), or clone F7.2.38 (Dako).

In some embodiments, the target protein may be a member of the CD28 family. In some embodiments, the target protein may be a member of the CD28 family such as PD-1 (CD279), LAG3 (CD223), ICOS (CD278), CTLA4 (CD152), BTLA (CD272) or CD28.

In some particular embodiments, the bispecific antibody or bispecific antigen binding fragment comprises an antigen binding domain capable of binding to CD3 or a CD3 polypeptide, and an antigen binding domain capable of binding to TIM-3 comprising at the CDRs, light and heavy chain variable domains or other TIM-3 binding fragment of clone A11 or clone B10 described herein.

In some embodiments, the bispecific antibody of the present invention may exhibit at least one of the following properties:

a) increases or enhances cell killing (e.g. T cell mediated cell killing) of TIM-3 expressing cells (antibody dependent cell-mediated cytotoxicity, ADCC), e.g. TIM-3 expressing acute myeloid leukemia cells;

b) increases or enhances cell killing (e.g. T cell mediated cell killing) of TIM-3 expressing stem cells (antibody dependent cell-mediated cytotoxicity, ADCC), e.g. TIM-3 expressing, CD34+ acute myeloid leukemia cells;

In some embodiments, the antigen binding domain for PD-1 may comprise the CDRs, light and heavy chain variable domains or other PD-1 binding fragment of e.g. anti-PD-1 antibody clone J116, clone MIH4 (eBioscience), clone 7A11B1 (Rockland Immunochemicals Inc.), clone 192106 (R&D Systems), clone J110, clone J105 (MBL International), clone 12A7D7, clone 7A11B1 (Abbiotec), clone #9X21 (MyBioSource.com), clone 4H4D1 (Proteintech Group), clone D3W4U, clone D3O4S (Cell Signaling Technology), clone RMP1-30, clone RMP1-14 (Merck Millipore), clone EH12.2H7 (BioLegend), clone 10B1227 (United States Biological), clone UMAB198, or clone UMAB197 (Origene Technologies). In some embodiments, the antigen binding domain for LAG3 may comprise the CDRs, light and heavy chain variable domains or other LAG3 binding fragment of e.g. anti-LAG3 antibody clone 17B4 (Enzo Life Sciences), clone 333210 (R&D Systems), or clone 14L676 (United States Biological). In some embodiments, the antigen binding domain for ICOS may comprise the CDRs, light and heavy chain variable domains or other ICOS binding fragment of e.g. anti-ICOS antibody clone ISA-3 (eBioscience), clone SP98 (Novus Biologicals), clone 1G1, clone 3G4 (Abnova Corporation), clone 669222 (R&D Systems), clone TQ09 (Creative Diagnostics), or clone C398.4A (BioLegend). In some embodiments, the antigen binding domain for CTLA4 may comprise the CDRs, light and heavy chain variable domains or other CTLA4 binding fragment of e.g. anti-CTLA4 antibody clone 2F1, clone 1F4 (Abnova Corporation), clone 9H10 (EMD Millipore), clone BNU3 (GeneTex), clone 1E2, clone AS32 (LifeSpan BioSciences) clone A3.4H2.H12 (Acris Antibodies), clone 060 (Sino

Biological), clone BU5G3 (Creative Diagnostics), clone MIH8 (MBL International), clone A3.6B10.G1, or clone L3D10 (BioLegend). In some embodiments, the antigen binding domain for BTLA may comprise the CDRs, light and heavy chain variable domains or other BTLA binding fragment of e.g. anti-BTLA antibody clone 1B7, clone 2G8, clone 4C5 (Abnova Corporation), clone 4B8 (antibodies-online), clone MIH26 (Thermo Scientific Pierce Antibodies), clone UMAB61 (OriGene Technologies), clone 330104 (R&D Systems), clone 1B4 (LifeSpan BioSciences), clone 440205, clone 5E7 (Creative Diagnostics). In some embodiments, the antigen binding domain for CD28 may comprise the CDRs, light and heavy chain variable domains or other CD28 binding fragment of e.g. anti-CD28 antibody clone CD28.6 (eBioscience), clone CD28.2, clone JJ319 (Novus Biologicals), clone 204.12, clone B-23, clone 10F3 (Thermo Scientific Pierce Antibodies), clone 37407 (R&D Systems), clone 204-12 (Abnova Corporation), clone 15E8 (EMD Millipore), clone 204-12, clone YTH913.12 (AbD Serotec), clone B-T3 (Acris Antibodies), clone 9H6E2 (Sino Biological), clone C28/77 (MyBioSource.com), clone KOLT-2 (ALPCO), clone 152-2E10 (Santa Cruz Biotechnology), or clone XPH-56 (Creative Diagnostics).

An antigen binding domain of a bispecific antibody or bispecific antigen binding fragment according to the present invention may be any domain of a polypeptide which is capable of binding to an antigen. In some embodiments, an antigen binding domain comprises at least the three light chain CDRs (i.e. LC-CDR1, LC-CDR2 and LC-CDR3) and three heavy chain CDRs (i.e. HC-CDR1, HC-CDR2 and HC-CDR3) which together define the antigen binding region of an antibody or antigen binding fragment. In some embodiments, an antigen binding domain may comprise the light chain variable domain and heavy chain variable domain of an antibody or antigen binding fragment. In some embodiments, an antigen binding domain may comprise the light chain polypeptide and heavy chain polypeptide of an antibody or antigen binding fragment.

Bispecific antibodies and bispecific antigen binding fragments according to the invention may be provided in any suitable format, such as those formats described in Kontermann MAbs 2012, 4(2): 182-197, which is hereby incorporated by reference in its entirety. For example, a bispecific antibody or bispecific antigen binding fragment may be a bispecific antibody conjugate (e.g. an IgG2, $F(ab')_2$ or CovX-Body), a bispecific IgG or IgG-like molecule (e.g. an IgG, $scFv_4$-Ig, IgG-scFv, scFv-IgG, DVD-Ig, IgG-sVD, sVD-IgG, 2 in 1-IgG, mAb^2, or Tandemab common LC), an asymmetric bispecific IgG or IgG-like molecule (e.g. a kih IgG, kih IgG common LC, CrossMab, kih IgG-scFab, mAb-Fv, charge pair or

SEED-body), a small bispecific antibody molecule (e.g. a Diabody (Db), dsDb, DART, scDb, tandAbs, tandem scFv (taFv), tandem dAb/VHH, triple body, triple head, Fab-scFv, or F(ab')$_2$-scFv$_2$), a bispecific Fc and C_H3 fusion protein (e.g. a taFv-Fc, Di-diabody, scDb-C_H3, scFv-Fc-scFv, HCAb-VHH, scFv-kih-Fc, or scFv-kih-C_H3), or a bispecific fusion protein (e.g. a scFv$_2$-albumin, scDb-albumin, taFv-toxin, DNL-Fab$_3$, DNL-Fab$_4$-IgG, DNL-Fab$_4$-IgG-cytokine$_2$). See in particular Figure 2 of Kontermann MAbs 2012, 4(2): 182-19.

The skilled person is able to design and prepare bispecific antibodies and bispecific antigen binding fragments according to the present invention.

Methods for producing bispecific antibodies include chemically crosslinking of antibodies or antibody fragments, e.g. with reducible disulphide or non-reducible thioether bonds, for example as described in Segal and Bast, 2001. Production of Bispecific Antibodies. Current Protocols in Immunology. 14:IV:2.13:2.13.1–2.13.16, which is hereby incorporated by reference in its entirety. For example, *N*-succinimidyl-3-(-2-pyridyldithio)-propionate (SPDP) can be used to chemically crosslink e.g. Fab fragments via hinge region SH- groups, to create disulfide-linked bispecific F(ab)$_2$ heterodimers.

Other methods for producing bispecific antibodies include fusing antibody-producing hybridomas e.g. with polyethylene glycol, to produce a quadroma cell capable of secreting bispecific antibody, for example as described in D. M. and Bast, B. J. 2001. Production of Bispecific Antibodies. Current Protocols in Immunology. 14:IV:2.13:2.13.1–2.13.16.

Bispecific antibodies and bispecific antigen binding fragments according to the present invention can also be produced recombinantly, by expression from e.g. a nucleic acid construct encoding polypeptides for the antigen binding molecules, for example as described in Antibody Engineering: Methods and Protocols, Second Edition (Humana Press, 2012), at Chapter 40: Production of Bispecific Antibodies: Diabodies and Tandem scFv (Hornig and Färber-Schwarz), or French, How to make bispecific antibodies, Methods Mol. Med. 2000; 40:333-339, the entire contents of both of which are hereby incorporated by reference.

For example, a DNA construct encoding the light and heavy chain variable domains for the two antigen binding domains (i.e. the light and heavy chain variable domains for the antigen binding domain capable of binding TIM-3, and the light and heavy chain variable

domains for the antigen binding domain capable of binding to another target protein), and including sequences encoding a suitable linker or dimerization domain between the antigen binding domains can be prepared by molecular cloning techniques. Recombinant bispecific antibody can thereafter be produced by expression (e.g. *in vitro*) of the construct in a suitable host cell (e.g. a mammalian host cell), and expressed recombinant bispecific antibody can then optionally be purified.

Antibodies, antigen fragments or polypeptides according to the present invention may also be used to construct chimeric antigen receptors (CAR; also called artificial T-cell receptors) in which a receptor is engineered by recombinant techniques to graft a selected specificity onto an immune cell. For example, the specificity of a monoclonal antibody may be grafted onto a T-cell, and the modified T-cells may find use in treatment of disease, e.g. cancer. One form of CAR is a fusion of an scFv comprising an antibody, antigen fragment or polypeptide according to the present invention to a transmembrane and endo domain of a suitable receptor scaffold. Techniques for the generation of CARs are described in Pule, M et al., (2003 *Cytotherapy* **5** (3): 211–26).

Antibodies may be produced by a process of affinity maturation in which a modified antibody is generated that has an improvement in the affinity of the antibody for antigen, compared to an unmodified parent antibody. Affinity-matured antibodies may be produced by procedures known in the art, e.g., Marks *et al.,Rio/Technology* 10:779-783 (1992); Barbas *et al. Proc Nat. Acad. Sci. USA* 91:3809-3813 (1994); Schier *et al. Gene* 169:147-155 (1995); Yelton *et al. J. Immunol.* 155:1994-2004 (1995); Jackson *et al., J. Immunol.* 154(7):331 0-15 9 (1995); and Hawkins *et al, J. Mol. Biol.* 226:889-896 (1992).

Antibodies according to the present invention preferably exhibit specific binding to TIM-3. An antibody that specifically binds to a target molecule preferably binds the target with greater affinity, and/or with greater duration than it binds to other targets. In one embodiment, the extent of binding of an antibody to an unrelated target is less than about 10% of the binding of the antibody to the target as measured, e.g., by ELISA, or by a radioimmunoassay (RIA). Alternatively, the binding specificity may be reflected in terms of binding affinity where the anti-TIM-3 antibody of the present invention binds to TIM-3 with a K_D that is at least 0.1 order of magnitude (i.e. 0.1 x 10^n, where n is an integer representing the order of magnitude) greater than the K_D of the antibody towards another target molecule, e.g. another member of the TIM-3 family. This may optionally be one of at least 0.2, 0.3, 0.4, 0.5, 0.6, 0.7, 0.8, 0.9, 1.0, 1.5, or 2.0.

Antibodies according to the present invention preferably have a dissociation constant (K_D) of one of ≤ 1μM, ≤ 100nM, ≤10nM, ≤1nM or ≤100pM. Binding affinity of an antibody for its target is often described in terms of its dissociation constant (K_D). Binding affinity can be measured by methods known in the art, such as by Surface Plasmon Resonance (SPR), or by a radiolabeled antigen binding assay (RIA) performed with the Fab version of the antibody and antigen molecule.

Antibodies according to the present invention may be "antagonist" antibodies that inhibit or reduce a biological activity of the antigen to which it binds. Blocking of TIM-3 assists in the restoration of T-cell function by inhibiting the immune-inhibitory signalling pathway mediated by TIM-3.

In some aspects, the antibody is clone A3, or a variant of A3. A3 comprises the following CDR sequences:

Light chain:

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

Heavy chain:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

CDR sequences determined by Kabat definition.

In some aspects, the antibody is clone B10, or a variant of B10. B10 comprises the following CDR sequences:

Light chain:

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

Heavy chain:

HC-CDR1:	GGSIGSSDYYWG	(SEQ ID NO:27), or
	SSDYYWG	(SEQ ID NO:59)

HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

CDR sequences determined by Kabat definition.

In some aspects, the antibody is clone G6, or a variant of G6. G6 comprises the following CDR sequences:

Light chain:

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

Heavy chain:

HC-CDR1:	GGSISSSNWWS	(SEQ ID NO:30), or
	SSNWWS	(SEQ ID NO:60)
HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

CDR sequences determined by Kabat definition.

In some aspects, the antibody is clone G7, or a variant of G7. G7 comprises the following CDR sequences:

Light chain:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

Heavy chain:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

CDR sequences determined by Kabat definition.

In some aspects, the antibody is clone G9, or a variant of G9. G9 comprises the following CDR sequences:

Light chain:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)

LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

Heavy chain:

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

CDR sequences determined by Kabat definition.

In some aspects, the antibody is clone A11 or clone A11_gl, or a variant of A11 or A11_gl. Each of A11 and A11_gl comprises the following CDR sequences:

Light chain:

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

Heavy chain:

HC-CDR1:	GGSFSGYYWS	(SEQ ID NO:52), or
	GYYWS	(SEQ ID NO:61)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

CDR sequences determined by Kabat definition.

Antibodies according to the present invention may comprise the CDRs of one of A3, B10, G6, G7, G9, A11, A11_gl or one of SEQ ID NOs 6, 7, 8, 24 or 58, 25, 26 or 9, 10, 11, 27 or 59, 28, 29 or 12, 13, 14, 30 or 60, 31, 32 or 15, 16, 17, 24 or 58, 25, 33 or 15, 16, 17, 24 or 58, 25, 34, or 47, 48, 49, 52 or 61, 53, 54, respectively. In an antibody according to the present invention one or two or three or four of the six CDR sequences may vary. A variant may have one or two amino acid substitutions in one or two of the six CDR sequences.

Amino acid sequences of the V_H and V_L chains of ant-TIM-3 clones are shown in Figures 1 and 2. The encoding nucleotide sequences are shown in Figure 4.

The light and heavy chain CDRs may also be particularly useful in conjunction with a number of different framework regions. Accordingly, light and/or heavy chains having LC-CDR1-3 or HC-CDR1-3 may possess an alternative framework region. Suitable framework regions are well known in the art and are described for example in M. Lefranc

& G. Le:franc (2001) "The Immunoglobulin FactsBook", Academic Press, incorporated herein by reference.

In this specification, antibodies may have V_H and/or VL chains comprising an amino acid sequence that has a high percentage sequence identity to one or more of the V_H and/or V_L amino acid sequences of SEQ ID NOs 1, 2, 3, 4, 5, 19, 20, 21, 22, 23, 45, 46, 50, 51 or to one or the amino acid sequences shown in Figure 1 and 2.

For example, antibodies according to the present invention include antibodies that bind TIM-3 and have a V_H or V_L chain that comprises an amino acid sequence having at least 70%, more preferably one of at least 75%, 80%, 85%, 86%, 87%, 88%, 89%, 90%, 91%, 92%, 93%, 94%, 95%, 96%, 97%, 98%, 99%, or 100%, sequence identity to the V_H or V_L chain amino acid sequence of one of SEQ ID NOs 1, 2, 3, 4, 5, 19, 20, 21, 22, 23, 45, 46, 50, 51 or to one or the amino acid sequences shown in Figures 1 and 2.

Antibodies according to the present invention may be detectably labelled or, at least, capable of detection. For example, the antibody may be labelled with a radioactive atom or a coloured molecule or a fluorescent molecule or a molecule which can be readily detected in any other way. Suitable detectable molecules include fluorescent proteins, luciferase, enzyme substrates, and radiolabels. The binding moiety may be directly labelled with a detectable label or it may be indirectly labelled. For example, the binding moiety may be an unlabelled antibody which can be detected by another antibody which is itself labelled. Alternatively, the second antibody may have bound to it biotin and binding of labelled streptavidin to the biotin is used to indirectly label the first antibody.

Methods of detection

Antibodies, or antigen binding fragments, described herein may be used in methods that involve the binding of the antibody or antigen binding fragment to TIM-3. Such methods may involve detection of the bound complex of antibody, or antigen binding fragment, and TIM-3. As such, in one embodiment a method is provided, the method comprising contacting a sample containing, or suspected to contain, TIM-3 with an antibody or antigen binding fragment as described herein and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.

Suitable method formats are well known in the art, including immunoassays such as sandwich assays, e.g. ELISA. The method may involve labelling the antibody, or antigen

binding fragment, or TIM-3, or both, with a detectable label, e.g. fluorescent, luminescent or radio- label.

Methods of this kind may provide the basis of a method of diagnosis of a disease or condition requiring detection and or quantitation of TIM-3. Such methods may be performed *in vitro* on a patient sample, or following processing of a patient sample. Once the sample is collected, the patient is not required to be present for the *in vitro* method of diagnosis to be performed and therefore the method may be one which is not practised on the human or animal body.

Such methods may involve determining the amount of TIM-3 present in a patient sample. The method may further comprise comparing the determined amount against a standard or reference value as part of the process of reaching a diagnosis. Other diagnostic tests may be used in conjunction with those described here to enhance the accuracy of the diagnosis or prognosis or to confirm a result obtained by using the tests described here.

The level of TIM-3 present in a patient sample may be indicative that a patient may respond to treatment with an anti-TIM-3 antibody. The presence of a high level of TIM-3 in a sample may be used to select a patient for treatment with an anti-TIM-3 antibody. The antibodies of the present invention may therefore be used to select a patient for treatment with anti-TIM-3 therapy.

Detection in a sample of TIM-3 may be used for the purpose of diagnosis of a T-cell dysfunctional disorder or a cancerous condition in the patient, diagnosis of a predisposition to a cancerous condition or for providing a prognosis (prognosticating) of a cancerous condition. The diagnosis or prognosis may relate to an existing (previously diagnosed) cancerous condition, which may be benign or malignant, may relate to a suspected cancerous condition or may relate to the screening for cancerous conditions in the patient (which may be previously undiagnosed).

In one embodiment the level of TIM-3 expression on CD8+ T cells may be detected in order to indicate the degree of T-cell exhaustion and severity of the disease state. In some cases, the level of TIM-3 expression on T cells or tumor cells may be used to select a patient for treatment with a modulator of TIM3 signalling, such as an anti-TIM3 antibody or anti-TIM3 agent.

A sample may be taken from any tissue or bodily fluid. The sample may comprise or may be derived from: a quantity of blood; a quantity of serum derived from the individual's blood which may comprise the fluid portion of the blood obtained after removal of the fibrin clot and blood cells; a tissue sample or biopsy; or cells isolated from said individual.

Methods according to the present invention are preferably performed *in vitro*. The term "*in vitro*" is intended to encompass experiments with cells in culture whereas the term "*in vivo*" is intended to encompass experiments with intact multi-cellular organisms.

Therapeutic applications

Antibodies, antigen binding fragments and polypeptides according to the present invention and compositions comprising such agents may be provided for use in methods of medical treatment. Treatment may be provided to subjects having a disease or condition in need of treatment. The disease or condition may be one of a T-cell dysfunctional disorder, including a T-cell dysfunctional disorder associated with a cancer, a cancer, or an infectious disease.

A T-cell dysfunctional disorder may be a disease or condition in which normal T-cell function is impaired causing downregulation of the subject's immune response to pathogenic antigens, e.g. generated by infection by exogenous agents such as microorganisms, bacteria and viruses, or generated by the host in some disease states such as in some forms of cancer (e.g. in the form of tumor associated antigens).

The T-cell dysfunctional disorder may comprise T-cell exhaustion or T-cell anergy. T-cell exhaustion comprises a state in which $CD8^+$ T-cells fail to proliferate or exert T-cell effector functions such as cytotoxicity and cytokine (e.g. IFNγ) secretion in response to antigen stimulation. Exhausted T-cells may also be characterised by sustained upregulation of TIM-3, where blockade of TIM-3:galectin 9 interactions may reverse the T-cell exhaustion and restore antigen-specific T cell responses.

The T-cell dysfunctional disorder may be manifest as an infection, or inability to mount an effective immune response against an infection. The infection may be chronic, persistent, latent or slow, and may be the result of bacterial, viral, fungal or parasitic infection. As such, treatment may be provided to patients having a bacterial, viral or fungal infection. Examples of bacterial infections include infection with *Helicobacter pylori*. Examples of viral infections include infection with HIV, hepatitis B or hepatitis C.

The T-cell dysfunctional disorder may be associated with a cancer, such as tumor immune escape. Many human tumors express tumor-associated antigens recognised by T cells and capable of inducing an immune response. However, immune evasion is common and is believed to be mediated by a number of soluble factors, including galectin 9. As such, blocking the interaction of TIM-3 and galectin 9 may inhibit this negative immunoregulatory signal to tumor cells and enhance tumor-specific $CD8^+$ T-cell immunity.

Cancers may also be treated where there is no indication of a T-cell dysfunctional disorder such as T-cell exhaustion. Antibodies according to the present invention may be cytotoxic against TIM-3 expressing cells, such as T-cells, e.g. exhausted T-cells or cancer cells such as acute myeloid leukemia cells. As such, the antibody, antigen binding fragment or polypeptides described herein may be useful in methods involving antibody dependent cell-mediated cytotoxicity (ADCC) or complement dependent cytotoxicity (CDC), or in any method that recruits immune effector functions to kill the target cells, such as CAR cells, or bispecific antibodies targeting CD3.

The use of an antibody, antigen binding fragment or polypeptide according to the present invention allows or may also allow the subject to suppress TIM-3 signalling and mount an effective immune response with limited impairment, evasion or induction of tumor immune escape. In such treatments, the antibody, antigen binding fragment or polypeptide may provide a treatment for cancer that involves prevention of the development of tumor immune escape.

The treatment may be aimed at prevention of the T-cell dysfunctional disorder, e.g. prevention of infection or of the development or progression of a cancer. As such, the antibodies, antigen binding fragments and polypeptides may be used to formulate pharmaceutical compositions or medicaments and subjects may be prophylactically treated against development of a disease state. This may take place before the onset of symptoms of the disease state, and/or may be given to subjects considered to be at greater risk of infection or development of cancer.

Treatment may comprise co-therapy with a vaccine, e.g. T-cell vaccine, which may involve simultaneous, separate or sequential therapy, or combined administration of vaccine and antibody, antigen binding fragment or polypeptide in a single composition. In this context, the antibody, antigen binding fragment or polypeptide may be provided as an

adjuvant to the vaccine. Limited proliferative potential of exhausted T cells has been attributed as a main reason for failure of T-cell immunotherapy and combination an agent capable of blocking or reversing T cell exhaustion is a potential strategy for improving the efficacy of T-cell immunotherapy (Barber et al., *Nature* Vol 439, No. 9 p682-687 Feb 2006).

Administration of an antibody, antigen binding fragment or polypeptide is preferably in a "therapeutically effective amount", this being sufficient to show benefit to the individual. The actual amount administered, and rate and time-course of administration, will depend on the nature and severity of the disease being treated. Prescription of treatment, e.g. decisions on dosage etc., is within the responsibility of general practitioners and other medical doctors, and typically takes account of the disorder to be treated, the condition of the individual patient, the site of delivery, the method of administration and other factors known to practitioners. Examples of the techniques and protocols mentioned above can be found in Remington's Pharmaceutical Sciences, 20th Edition, 2000, pub. Lippincott, Williams & Wilkins.

Formulating pharmaceutically useful compositions and medicaments

Antibodies, antigen binding fragments and polypeptides according to the present invention may be formulated as pharmaceutical compositions for clinical use and may comprise a pharmaceutically acceptable carrier, diluent, excipient or adjuvant.

In accordance with the present invention methods are also provided for the production of pharmaceutically useful compositions, such methods of production may comprise one or more steps selected from: isolating an antibody, antigen binding fragment or polypeptide as described herein; and/or mixing an isolated antibody, antigen binding fragment or polypeptide as described herein with a pharmaceutically acceptable carrier, adjuvant, excipient or diluent.

For example, a further aspect of the present invention relates to a method of formulating or producing a medicament or pharmaceutical composition for use in the treatment of a T-cell dysfunctional disorder, the method comprising formulating a pharmaceutical composition or medicament by mixing an antibody, antigen binding fragment or polypeptide as described herein with a pharmaceutically acceptable carrier, adjuvant, excipient or diluent.

Infection

An infection may be any infection or infectious disease, e.g. bacterial, viral, fungal, or parasitic infection. In some embodiments it may be particularly desirable to treat chronic/persistent infections, e.g. where such infections are associated with T cell dysfunction or T cell exhaustion.

It is well established that T cell exhaustion is a state of T cell dysfunction that arises during many chronic infections (including viral, bacterial and parasitic), as well as in cancer (Wherry *Nature Immunology* Vol.12, No.6, p492-499, June 2011).

TIM-3 expression has been reported to play an important pathogenic role in patients having chronic infection (e.g. as reported by Golden-Mason L, et al., J Virol. 2009;83(18):9122–9130.)

Examples of bacterial infections that may be treated include infection by *Bacillus spp., Bordetella pertussis, Clostridium spp., Corynebacterium spp., Vibrio chloerae, Staphylococcus spp., Streptococcus spp. Escherichia, Klebsiella, Proteus, Yersinia, Erwina, Salmonella, Listeria* sp, *Helicobacter pylori*, mycobacteria (e.g. *Mycobacterium tuberculosis*) and *Pseudomonas aeruginosa.* For example, the bacterial infection may be sepsis or tuberculosis.

Yao et al (PD-1 on dendritic cells impedes innate immunity against bacterial infection. *Blood* 113(23):5811-5818 Jun 4 2009) established PD-1 in the negative regulation of DC function during innate immune response to infection by *Listeria monocytogenes*. Brahmamdam et al (Delayed administration of anti-PD-1 antibody reverses immune dysfunction and improves survival during sepsis. *Journal of Leukocyte Biology* vo.88, no.2 233-240, August 2010) reported that anti-PD-1 antibody administered 24 h after sepsis prevented sepsis-induced depletion of lymphocytes and DCs, increased Bcl-xL, blocked apoptosis and improved survival. Tim3:Galectin-9 interactions have been reported to mediate T cell exhaustion and mediate the innate and adaptive immune response to infection by Mycobacterium tuberculosis (Jayaraman et al., *The Journal of Immunology* 2012, 188, 70.6).

Examples of viral infections that may be treated include infection by influenza virus, measles virus, hepatitis B virus (HBV), hepatitis C virus (HCV), human immunodeficiency

virus (HIV), lymphocytic choriomeningitis virus (LCMV), Herpes simplex virus and human papilloma virus.

Chronic viral infections, such as those caused by HCV, HBV, and HIV commonly involve mechanisms to evade immune clearance. Expression of PD-1 and TIM-3 have been identified as correlating with defective T cell responses to hepatitis C virus (HCV) (McMahan et al., The Journal of Clinical Investigation Vol. 120, No. 12 p4546-4557, December 2010). In HCV, McMahan et al (*supra*) found that the level of dual TIM-3 and PD-1 expression on HCV-specific CTLs predated the development of viral persistence, providing prognostic information. Barber et al. (Nature Vol 439, No. 9 p682-687 Feb 2006) reported that PD-1 is upregulated during chronic viral infection. In mice infected with LCMV they reported that blockade of the PD-1/PD-L1 inhibitory pathway had a beneficial effect on CD8 T cells, restoring their ability to undergo proliferation, secrete cytokines, kill infected cells and decrease viral load. PD-1 is also upregulated in HIV infection (Said et al., *Nature Medicine* Vol. 16, No.4 p452-460 April 2010). Blocking interaction between PD-1 and PD-L1 contributed to viral clearance and improved T cell function in animal models of chronic viral infection (Said et al., *supra*).

Examples of fungal infections that may be treated include infection by *Alternaria sp, Aspergillus sp, Candida sp* and *Histoplasma sp*. The fungal infection may be fungal sepsis or histoplasmosis.

Chang et al (Blockade of the negative co-stimulatory molecules PD-1 and CTLA-4 improves survival in primary and secondary fungal sepsis. *Critical Care* 2013, 17:R85) reported that anti-PD1 antibodies were highly effective at improving survival in primary and secondary fungal sepsis. Lázár-Molnár et al (The PD-1/PD-L costimulatory pathway critically affects host resistance to the pathogenic fungus *Histoplasma capsulatum* PNAS vol. 105, no.7, p2658-2663, 19 Feb 2008) reported that anti-PD-1 antibody significantly increased survival of mice infected with *Histoplasma capsulatum*. As such, the importance of T cell exhaustion in mediating fungal infection is well established. Examples of parasitic infections that may be treated include infection by *Plasmodium* species (e.g. *Plasmodium falciparum, Plasmodium yoeli*, *Plasmodium ovale*, *Plasmodium* vivax, or *Plasmodium chabaudi chabaudi*). The parasitic infection may be a disease such as malaria, leishmaniasis and toxoplasmosis.

Infection of humans with *Plasmodium falciparum* has been shown to result in higher expression of PD-1 and T cell exhaustion mice (Butler et al., *Nature Immunology* Vol.13, No.12, p 188-195 February 2012). Blockade of PD-L1 and LAG-3 using anti-PD-L1 and anti-LAG-3 monoclonal antibodies *in vivo* contributed to the restoration of $CD4^+$ T-cell function, amplification of the number of follicular helper T cells, germinal-center B cells and plasmablasts, enhanced protective antibodies and rapidly cleared blood-stage malaria in mice. It was also shown to block the development of chronic infection (Butler et al.,*supra*).

Cancer

A cancer may be any unwanted cell proliferation (or any disease manifesting itself by unwanted cell proliferation), neoplasm or tumor or increased risk of or predisposition to the unwanted cell proliferation, neoplasm or tumor. The cancer may be benign or malignant and may be primary or secondary (metastatic). A neoplasm or tumor may be any abnormal growth or proliferation of cells and may be located in any tissue. Examples of tissues include the adrenal gland, adrenal medulla, anus, appendix, bladder, blood, bone, bone marrow, brain, breast, cecum, central nervous system (including or excluding the brain) cerebellum, cervix, colon, duodenum, endometrium, epithelial cells (e.g. renal epithelia), gallbladder, oesophagus, glial cells, heart, ileum, jejunum, kidney, lacrimal glad, larynx, liver, lung, lymph, lymph node, lymphoblast, maxilla, mediastinum, mesentery, myometrium, nasopharynx, omentume, oral cavity, ovary, pancreas, parotid gland, peripheral nervous system, peritoneum, pleura, prostate, salivary gland, sigmoid colon, skin, small intestine, soft tissues, spleen, stomach, testis, thymus, thyroid gland, tongue, tonsil, trachea, uterus, vulva, white blood cells.

Tumors to be treated may be nervous or non-nervous system tumors. Nervous system tumors may originate either in the central or peripheral nervous system, e.g. glioma, medulloblastoma, meningioma, neurofibroma, ependymoma, Schwannoma, neurofibrosarcoma, astrocytoma and oligodendroglioma. Non-nervous system cancers/tumors may originate in any other non-nervous tissue, examples include melanoma, mesothelioma, lymphoma, myeloma, leukemia, Non-Hodgkin's lymphoma (NHL), Hodgkin's lymphoma, chronic myelogenous leukemia (CML), acute myeloid leukemia (AML), myelodysplastic syndrome (MDS), cutaneous T-cell lymphoma (CTCL), chronic lymphocytic leukemia (CLL), hepatoma, epidermoid carcinoma, prostate carcinoma, breast cancer, lung cancer , colon cancer, ovarian cancer, pancreatic cancer, thymic carcinoma, NSCLC, haematologic cancer and sarcoma.

Adoptive T cell transfer therapy

Adoptive T cell transfer therapy generally refers to a process in which white blood cells are removed from a subject, typically by drawing a blood sample from which white blood cells are separated, expanded *in vitro* or *ex vivo* and returned either to the same subject or to a different subject. The treatment is typically aimed at increasing the amount/concentration of an active form of the required T cell population in the subject. Such treatment may be beneficial in subjects experiencing T cell exhaustion.

Antibodies capable of blocking the mechanism of T cell exhaustion, or reversing it, provide a means of enhancing T cell activity and promoting T cell expansion.

Accordingly, in a further aspect of the present invention a method is provided for expanding a population of T cells, wherein T cells are contacted *in vitro* or *ex vivo* with an antibody, antigen binding fragment or polypeptide according to the present invention.

The method may optionally comprise one or more of the following steps: taking a blood sample from a subject; isolating T cells from the blood sample; culturing the T cells in *in vitro* or *ex vivo* cell culture (where they may be contacted with the antibody, antigen binding fragment or polypeptide), collecting an expanded population of T cells; mixing the T cells with an adjuvant, diluent, or carrier; administering the expanded T cells to a subject.

Accordingly, in some aspects of the present invention a method of treatment of a subject having a T-cell dysfunctional disorder is provided, the method comprising obtaining a blood sample from a subject in need of treatment, culturing T cells obtained from the blood sample in the presence of an antibody, antigen binding fragment or polypeptide according to the present invention so as to expand the T cell population, collecting expanded T cells, and administering the expanded T cells to a subject in need of treatment.

The T cells may be obtained from a subject requiring treatment, and may be isolated and/or purified. They may be a $CD4^+$ and/or $CD8^+$ T-cell population. The T-cells may represent a population experiencing T cell exhaustion and may optionally have upregulated expression of TIM-3.

During culture, T cells may be contacted with the antibody, antigen binding fragment or polypeptide under conditions and for a period of time suitable to allow expansion of the T cells to a desired number of cells. After a suitable period of time the T cells may be harvested, optionally concentrated, and may be mixed with a suitable carrier, adjuvant or diluent and returned to the subject's body. A subject may undergo one or more rounds of such therapy.

Methods of T cell expansion are well known in the art, such as those described in Kalamasz et al., *J Immunother* 2004 Sep-Oct; 27(5):405-18; Montes et al., *Clin Exp Immunol* 2005 Nov;142(2):292-302; Wölfl and Greenburg *Nature Protocols* 9 p950-966 27 March 2014; Trickett and Kwan *Journal of Immunological Methods* Vol. 275, Issues 1-2, 1 April 2003, p251-255; Butler et al *PLoSONE* 7(1) 12 Jan 2012.

Simultaneous or Sequential Administration

Compositions may be administered alone or in combination with other treatments, either simultaneously or sequentially dependent upon the condition to be treated.

In this specification an antibody, antigen binding fragment or polypeptide of the present invention and an anti-infective agent or chemotherapeutic agent (therapeutic agent) may be administered simultaneously or sequentially.

In some embodiments, treatment with an antibody, antigen binding fragment or polypeptide of the present invention may be accompanied by chemotherapy.

Simultaneous administration refers to administration of the antibody, antigen binding fragment or polypeptide and therapeutic agent together, for example as a pharmaceutical composition containing both agents (combined preparation), or immediately after each other and optionally via the same route of administration, e.g. to the same artery, vein or other blood vessel.

Sequential administration refers to administration of one of the antibody, antigen binding fragment or polypeptide or therapeutic agent followed after a given time interval by separate administration of the other agent. It is not required that the two agents are administered by the same route, although this is the case in some embodiments. The time interval may be any time interval.

Anti-infective agents

In treating infection, an antibody, antigen binding fragment or polypeptide of the present invention may be administered in combination with an anti-infective agent, as described above. The anti-infective agent may be an agent known to have action against the microorganism or virus responsible for the infection.

Suitable anti-infective agents include antibiotics (such as penicillins, cephalosporins, rifamycins, lipiarmycins, quinolones, sulfonamides, macrolides, lincosamides, tetracyclines, cyclic lipopeptides, glycylcyclines, oxazolidinones, and lipiarmycins), anti-viral agents (such as reverse transcriptase inhibitors, integrase inhibitors, transcription factor inhibitors, antisense and siRNA agents and protease inhibitors), anti-fungal agents (such as polyenes, imidiazoles, triazoles, thiazoles, allylamines, and echinocandins) and anti-parasitic agents (such as antinematode agents, anticestode agents, antitrematode agents, antiamoebic agents and antiprotozoal agents).

Chemotherapy

Chemotherapy refers to treatment of a cancer with a drug or with ionising radiation (e.g. radiotherapy using X-rays or γ-rays). In preferred embodiments chemotherapy refers to treatment with a drug. The drug may be a chemical entity, e.g. small molecule pharmaceutical, antibiotic, DNA intercalator, protein inhibitor (e.g. kinase inhibitor), or a biological agent, e.g. antibody, antibody fragment, nucleic acid or peptide aptamer, nucleic acid (e.g. DNA, RNA), peptide, polypeptide, or protein. The drug may be formulated as a pharmaceutical composition or medicament. The formulation may comprise one or more drugs (e.g. one or more active agents) together with one or more pharmaceutically acceptable diluents, excipients or carriers.

A treatment may involve administration of more than one drug. A drug may be administered alone or in combination with other treatments, either simultaneously or sequentially dependent upon the condition to be treated. For example, the chemotherapy may be a co-therapy involving administration of two drugs, one or more of which may be intended to treat the cancer.

The chemotherapy may be administered by one or more routes of administration, e.g. parenteral, intravenous injection, oral, subcutaneous, intradermal or intratumoral.

The chemotherapy may be administered according to a treatment regime. The treatment regime may be a pre-determined timetable, plan, scheme or schedule of chemotherapy administration which may be prepared by a physician or medical practitioner and may be tailored to suit the patient requiring treatment.

The treatment regime may indicate one or more of: the type of chemotherapy to administer to the patient; the dose of each drug or radiation; the time interval between administrations; the length of each treatment; the number and nature of any treatment holidays, if any etc. For a co-therapy a single treatment regime may be provided which indicates how each drug is to be administered.

Chemotherapeutic drugs and biologics may be selected from:

- alkylating agents such as cisplatin, carboplatin, mechlorethamine, cyclophosphamide, chlorambucil, ifosfamide;
- purine or pyrimidine anti-metabolites such as azathiopurine or mercaptopurine;
- alkaloids and terpenoids, such as vinca alkaloids (e.g. vincristine, vinblastine, vinorelbine, vindesine), podophyllotoxin, etoposide, teniposide, taxanes such as paclitaxel (Taxol™), docetaxel;
- topoisomerase inhibitors such as the type I topoisomerase inhibitors camptothecins irinotecan and topotecan, or the type II topoisomerase inhibitors amsacrine, etoposide, etoposide phosphate, teniposide;
- antitumor antibiotics (e.g. anthracyline antibiotics) such as dactinomycin, doxorubicin (Adriamycin™), epirubicin, bleomycin, rapamycin;
- antibody based agents, such as anti-PD-1 antibodies, anti-PD-L1, anti-CTLA-4, anti-LAG-3, anti-4-1BB, anti-GITR, anti-CD27, anti-BLTA, anti-OX40, anti-VEGF, anti-TNFα, anti-IL-2, antiGpIIb/IIIa, anti-CD-52, anti-CD20, anti-RSV, anti-HER2/neu(erbB2), anti-TNF receptor, anti-EGFR antibodies, monoclonal antibodies or antibody fragments, examples include: cetuximab, panitumumab, infliximab, basiliximab, bevacizumab (Avastin®), abciximab, daclizumab, gemtuzumab, alemtuzumab, rituximab (Mabthera®), palivizumab, trastuzumab, etanercept, adalimumab, nimotuzumab

- EGFR inihibitors such as erlotinib, cetuximab and gefitinib
- anti-angiogenic agents such as bevacizumab (Avastin®)
- anti-cancer vaccines such as Sipuleucel-T (Provenge®)

In one embodiment the chemotherapeutic agent is an anti-PD-1 or an anti-PD-L1, anti-CTLA-4, anti-LAG-3, anti-4-1BB, anti-GITR, anti-CD27, anti-BLTA, anti-OX40, anti-VEGF, anti-TNF-α, anti-IL-2, anti-GpIIb/IIIa, anti-CD52, anti-CD20, anti-RSV, anti-HER2/neu(erb2), anti-TNF receptor, anti-EGFR antibody. In some embodiments, the chemotherapeutic agent is an immune checkpoint inhibitor or costimulation molecule.

Further chemotherapeutic drugs may be selected from: 13-cis-Retinoic Acid, 2-Chlorodeoxyadenosine, 5-Azacitidine 5-Fluorouracil, 6-Mercaptopurine, 6-Thioguanine, Abraxane, Accutane®, Actinomycin-D Adriamycin®, Adrucil®, Afinitor®, Agrylin®, Ala-Cort®, Aldesleukin, Alemtuzumab, ALIMTA, Alitretinoin, Alkaban-AQ®, Alkeran®, All-transretinoic Acid, Alpha Interferon, Altretamine, Amethopterin, Amifostine, Aminoglutethimide, Anagrelide, Anandron®, Anastrozole, Arabinosylcytosine, Aranesp®, Aredia®, Arimidex®, Aromasin®, Arranon®, Arsenic Trioxide, Asparaginase, ATRA Avastin®, Azacitidine, BCG, BCNU, Bendamustine, Bevacizumab, Bexarotene, BEXXAR®, Bicalutamide, BiCNU, Blenoxane®, Bleomycin, Bortezomib, Busulfan, Busulfex®, Calcium Leucovorin, Campath®, Camptosar®, Camptothecin-11, Capecitabine, Carac™, Carboplatin, Carmustine, Casodex®, CC-5013, CCI-779, CCNU, CDDP, CeeNU, Cerubidine®, Cetuximab, Chlorambucil, Cisplatin, Citrovorum Factor, Cladribine, Cortisone, Cosmegen®, CPT-11, Cyclophosphamide, Cytadren®, Cytarabine Cytosar-U®, Cytoxan®, Dacogen, Dactinomycin, Darbepoetin Alfa, Dasatinib, Daunomycin, Daunorubicin, Daunorubicin Hydrochloride, Daunorubicin Liposomal, DaunoXome®, Decadron, Decitabine, Delta-Cortef®, Deltasone®, Denileukin, Diftitox, DepoCyt™, Dexamethasone, Dexamethasone Acetate, Dexamethasone Sodium Phosphate, Dexasone, Dexrazoxane, DHAD, DIC, Diodex, Docetaxel, Doxil®, Doxorubicin, Doxorubicin Liposomal, Droxia™, DTIC, DTIC-Dome®, Duralone®, Eligard™, Ellence™, Eloxatin™, Elspar®, Emcyt®, Epirubicin, Epoetin Alfa, Erbitux, Erlotinib, Erwinia L-asparaginase, Estramustine, Ethyol Etopophos®, Etoposide, Etoposide Phosphate, Eulexin®, Everolimus, Evista®, Exemestane, Faslodex®, Femara®, Filgrastim, Floxuridine, Fludara®, Fludarabine, Fluoroplex®, Fluorouracil, Fluoxymesterone, Flutamide, Folinic Acid, FUDR®, Fulvestrant, Gefitinib, Gemcitabine, Gemtuzumab ozogamicin, Gleevec™, Gliadel® Wafer, Goserelin, Granulocyte - Colony Stimulating Factor, Granulocyte Macrophage Colony Stimulating Factor, Herceptin ®, Hexadrol, Hexalen®, Hexamethylmelamine, HMM, Hycamtin®, Hydrea®, Hydrocort Acetate®, Hydrocortisone, Hydrocortisone Sodium Phosphate, Hydrocortisone Sodium Succinate, Hydrocortone Phosphate, Hydroxyurea, Ibritumomab, Ibritumomab Tiuxetan, Idamycin®, Idarubicin, Ifex®, IFN-alpha, Ifosfamide, IL-11, IL-2, Imatinib mesylate,

Imidazole Carboxamide, Interferon alfa, Interferon Alfa-2b (PEG Conjugate), Interleukin - 2, Interleukin-11, Intron A® (interferon alfa-2b), Iressa®, Irinotecan, Isotretinoin, Ixabepilone, Ixempra™, Kidrolase, Lanacort®, Lapatinib, L-asparaginase, LCR, Lenalidomide, Letrozole, Leucovorin, Leukeran, Leukine™, Leuprolide, Leurocristine, Leustatin™, Liposomal Ara-C, Liquid Pred®, Lomustine, L-PAM, L-Sarcolysin, Lupron®, Lupron Depot®, Matulane®, Maxidex, Mechlorethamine, Mechlorethamine Hydrochloride, Medralone®, Medrol®, Megace®, Megestrol, Megestrol Acetate, Melphalan, Mercaptopurine, Mesna, Mesnex™, Methotrexate, Methotrexate Sodium, Methylprednisolone, Meticorten®, Mitomycin, Mitomycin-C, Mitoxantrone, M-Prednisol®, MTC, MTX, Mustargen®, Mustine, Mutamycin®, Myleran®, Mylocel™, Mylotarg®, Navelbine®, Nelarabine, Neosar®, Neulasta™, Neumega®, Neupogen®, Nexavar®, Nilandron®, Nilutamide, Nipent®, Nitrogen Mustard, Novaldex®, Novantrone®, Octreotide, Octreotide acetate, Oncospar®, Oncovin®, Ontak®, Onxal™, Oprevelkin, Orapred®, Orasone®, Oxaliplatin, Paclitaxel, Paclitaxel Protein-bound, Pamidronate, Panitumumab, Panretin®, Paraplatin®, Pediapred®, PEG Interferon, Pegaspargase, Pegfilgrastim, PEG-INTRON™, PEG-L-asparaginase, PEMETREXED, Pentostatin, Phenylalanine Mustard, Platinol®, Platinol-AQ®, Prednisolone, Prednisone, Prelone®, Procarbazine, PROCRIT®, Proleukin®, Prolifeprospan 20 with Carmustine Implant Purinethol®, Raloxifene, Revlimid®, Rheumatrex®, Rituxan®, Rituximab, Roferon-A® (Interferon Alfa-2a), Rubex®, Rubidomycin hydrochloride, Sandostatin® Sandostatin LAR®, Sargramostim, Solu-Cortef®, Solu-Medrol®, Sorafenib, SPRYCEL™, STI-571, Streptozocin, SU11248, Sunitinib, Sutent®, Tamoxifen, Tarceva®, Targretin®, Taxol®, Taxotere®, Temodar®, Temozolomide, Temsirolimus, Teniposide, TESPA, Thalidomide, Thalomid®, TheraCys®, Thioguanine, Thioguanine Tabloid®, Thiophosphoamide, Thioplex®, Thiotepa, TICE®, Toposar®, Topotecan, Toremifene, Torisel®, Tositumomab, Trastuzumab, Treanda®, Tretinoin, Trexall™, Trisenox®, TSPA, TYKERB®, VCR, Vectibix™, Velban®, Velcade®, VePesid®, Vesanoid®, Viadur™, Vidaza®, Vinblastine, Vinblastine Sulfate, Vincasar Pfs®, Vincristine, Vinorelbine, Vinorelbine tartrate, VLB, VM-26, Vorinostat, VP-16, Vumon®, Xeloda®, Zanosar®, Zevalin™, Zinecard®, Zoladex®, Zoledronic acid, Zolinza, Zometa®.

Routes of administration

Antibodies, antigen binding fragments, polypeptides and other therapeutic agents, medicaments and pharmaceutical compositions according to aspects of the present invention may be formulated for administration by a number of routes, including but not limited to, parenteral, intravenous, intra-arterial, intramuscular, subcutaneous,

intradermal, intratumoral and oral. Antibodies, antigen binding fragments, polypeptides and other therapeutic agents, may be formulated in fluid or solid form. Fluid formulations may be formulated for administration by injection to a selected region of the human or animal body.

Dosage regime

Multiple doses of the antibody, antigen binding fragment or polypeptide may be provided. One or more, or each, of the doses may be accompanied by simultaneous or sequential administration of another therapeutic agent.

Multiple doses may be separated by a predetermined time interval, which may be selected to be one of 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, or 31 days, or 1, 2, 3, 4, 5, or 6 months. By way of example, doses may be given once every 7, 14, 21 or 28 days (plus or minus 3, 2, or 1 days).

Kits

In some aspects of the present invention a kit of parts is provided. In some embodiments the kit may have at least one container having a predetermined quantity of the antibody, antigen binding fragment or polypeptide. The kit may provide the antibody, antigen binding fragment or polypeptide in the form of a medicament or pharmaceutical composition, and may be provided together with instructions for administration to a patient in order to treat a specified disease or condition. The antibody, antigen binding fragment or polypeptide may be formulated so as to be suitable for injection or infusion to a tumor or to the blood.

In some embodiments the kit may further comprise at least one container having a predetermined quantity of another therapeutic agent (e.g. anti-infective agent or chemotherapy agent). In such embodiments, the kit may also comprise a second medicament or pharmaceutical composition such that the two medicaments or pharmaceutical compositions may be administered simultaneously or separately such that they provide a combined treatment for the specific disease or condition. The therapeutic agent may also be formulated so as to be suitable for injection or infusion to a tumor or to the blood.

Subjects

The subject to be treated may be any animal or human. The subject is preferably mammalian, more preferably human. The subject may be a non-human mammal, but is more preferably human. The subject may be male or female. The subject may be a patient. A subject may have been diagnosed with a disease or condition requiring treatment, or be suspected of having such a disease or condition.

Protein Expression

Molecular biology techniques suitable for the producing polypeptides according to the invention in cells are well known in the art, such as those set out in Sambrook et al., Molecular Cloning: A Laboratory Manual, New York: Cold Spring Harbor Press, 1989

The polypeptide may be expressed from a nucleotide sequence. The nucleotide sequence may be contained in a vector present in a cell, or may be incorporated into the genome of the cell.

A "vector" as used herein is an oligonucleotide molecule (DNA or RNA) used as a vehicle to transfer exogenous genetic material into a cell. The vector may be an expression vector for expression of the genetic material in the cell. Such vectors may include a promoter sequence operably linked to the nucleotide sequence encoding the gene sequence to be expressed. A vector may also include a termination codon and expression enhancers. Any suitable vectors, promoters, enhancers and termination codons known in the art may be used to express polypeptides from a vector according to the invention. Suitable vectors include plasmids, binary vectors, viral vectors and artificial chromosomes (e.g. yeast artificial chromosomes).

In this specification the term "operably linked" may include the situation where a selected nucleotide sequence and regulatory nucleotide sequence (e.g. promoter and/or enhancer) are covalently linked in such a way as to place the expression of the nucleotide sequence under the influence or control of the regulatory sequence (thereby forming an expression cassette). Thus a regulatory sequence is operably linked to the selected nucleotide sequence if the regulatory sequence is capable of effecting transcription of the nucleotide sequence. Where appropriate, the resulting transcript may then be translated into a desired protein or polypeptide.

Any cell suitable for the expression of polypeptides may be used for producing peptides according to the invention. The cell may be a prokaryote or eukaryote. Suitable

prokaryotic cells include *E.coli*. Examples of eukaryotic cells include a yeast cell, a plant cell, insect cell or a mammalian cell. In some cases the cell is not a prokaryotic cell because some prokaryotic cells do not allow for the same post-translational modifications as eukaryotes. In addition, very high expression levels are possible in eukaryotes and proteins can be easier to purify from eukaryotes using appropriate tags. Specific plasmids may also be utilised which enhance secretion of the protein into the media.

Methods of producing a polypeptide of interest may involve culture or fermentation of a cell modified to express the polypeptide. The culture or fermentation may be performed in a bioreactor provided with an appropriate supply of nutrients, air/oxygen and/or growth factors. Secreted proteins can be collected by partitioning culture media/fermentation broth from the cells, extracting the protein content, and separating individual proteins to isolate secreted polypeptide. Culture, fermentation and separation techniques are well known to those of skill in the art.

Bioreactors include one or more vessels in which cells may be cultured. Culture in the bioreactor may occur continuously, with a continuous flow of reactants into, and a continuous flow of cultured cells from, the reactor. Alternatively, the culture may occur in batches. The bioreactor monitors and controls environmental conditions such as pH, oxygen, flow rates into and out of, and agitation within the vessel such that optimum conditions are provided for the cells being cultured.

Following culture of cells that express the polypeptide of interest, that polypeptide is preferably isolated. Any suitable method for separating polypeptides/proteins from cell culture known in the art may be used. In order to isolate a polypeptide/protein of interest from a culture, it may be necessary to first separate the cultured cells from media containing the polypeptide/protein of interest. If the polypeptide/protein of interest is secreted from the cells, the cells may be separated from the culture media that contains the secreted polypeptide/protein by centrifugation. If the polypeptide/protein of interest collects within the cell, it will be necessary to disrupt the cells prior to centrifugation, for example using sonification, rapid freeze-thaw or osmotic lysis. Centrifugation will produce a pellet containing the cultured cells, or cell debris of the cultured cells, and a supernatant containing culture medium and the polypeptide/protein of interest.

It may then be desirable to isolate the polypeptide/protein of interest from the supernatant or culture medium, which may contain other protein and non-protein components. A

common approach to separating polypeptide/protein components from a supernatant or culture medium is by precipitation. Polypeptides/proteins of different solubility are precipitated at different concentrations of precipitating agent such as ammonium sulfate. For example, at low concentrations of precipitating agent, water soluble proteins are extracted. Thus, by adding increasing concentrations of precipitating agent, proteins of different solubility may be distinguished. Dialysis may be subsequently used to remove ammonium sulfate from the separated proteins.

Other methods for distinguishing different polypeptides/proteins are known in the art, for example ion exchange chromatography and size chromatography. These may be used as an alternative to precipitation, or may be performed subsequently to precipitation.

Once the polypeptide/protein of interest has been isolated from culture it may be necessary to concentrate the protein. A number of methods for concentrating a protein of interest are known in the art, such as ultrafiltration or lyophilisation.

Sequence Identity

Alignment for purposes of determining percent amino acid or nucleotide sequence identity can be achieved in various ways known to a person of skill in the art, for instance, using publicly available computer software such as ClustalW 1.82. T-coffee or Megalign (DNASTAR) software. When using such software, the default parameters, e.g. for gap penalty and extension penalty, are preferably used. The default parameters of ClustalW 1.82 are: Protein Gap Open Penalty = 10.0, Protein Gap Extension Penalty = 0.2, Protein matrix = Gonnet, Protein/DNA ENDGAP = -1, Protein/DNA GAPDIST = 4.

The invention includes the combination of the aspects and preferred features described except where such a combination is clearly impermissible or expressly avoided.

The section headings used herein are for organizational purposes only and are not to be construed as limiting the subject matter described.

Aspects and embodiments of the present invention will now be illustrated, by way of example, with reference to the accompanying figures. Further aspects and embodiments will be apparent to those skilled in the art. All documents mentioned in this text are incorporated herein by reference.

Throughout this specification, including the claims which follow, unless the context requires otherwise, the word "comprise," and variations such as "comprises" and "comprising," will be understood to imply the inclusion of a stated integer or step or group of integers or steps but not the exclusion of any other integer or step or group of integers or steps.

It must be noted that, as used in the specification and the appended claims, the singular forms "a," "an," and "the" include plural referents unless the context clearly dictates otherwise. Ranges may be expressed herein as from "about" one particular value, and/or to "about" another particular value. When such a range is expressed, another embodiment includes from the one particular value and/or to the other particular value. Similarly, when values are expressed as approximations, by the use of the antecedent "about," it will be understood that the particular value forms another embodiment.

Brief Description of the Figures

Embodiments and experiments illustrating the principles of the invention will now be discussed with reference to the accompanying figures in which:

Figure 1. Light chain variable domain sequences for anti-TIM-3 antibody clones A3, B10, G6, G7, G9, A11 and A11_gl (human IgG4). CDRs are underlined and shown separately.

Figure 2. Heavy chain variable domain sequences for anti- TIM-3 antibody clones A3, B10, G6, G7, G9, A11 and A11_gl (human IgG4). CDRs are underlined and shown separately.

Figure 3. Table showing light chain and heavy chain CDR sequences for anti-TIM-3 antibody clones A3, B10, G6, G7, G9, A11 and A11_gl.

Figure 4. Nucleotide and encoded amino acid sequences of heavy and light chain variable domain sequences for anti-TIM-3 antibody clones A3, B10, G6, G7, G9, A11 and A11_gl.

Figure 5. Chart showing binding of clones A3, B10, G6, G7, and G9 to human and murine TIM-3 as determined by ELISA.

Figure 6. Chart showing blocking of human TIM-3:human galectin 9 interaction at the surface of MOLT3 cells by clones A3, B10, G6, G7, and G9.

Figure 7. Table showing affinity of clones A3, B10, G6, G7, and G9 for human TIM-3.

Figure 8. Charts showing cytotoxic effect of clones A3, B10, G6, G7, and G9 on acute myeloid leukemia cells OCI-AML3 (M4), and THP-1 (M5).

Figure 9. Chart showing binding of clone A11 to human and murine TIM-3.

Figure 10. Chart showing blocking of human TIM-3:human galectin 9 interaction at the surface of MOLT3 cells by clone A11 as determined by ELISA.

Figure 11. Table showing affinity of clone A11 for human TIM-3.

Figure 12. Chart showing cytotoxic effect of clone A11 on acute myeloid leukemia cells OCI-AML3 (M4), and THP-1 (M5).

Figure 13. Chart showing effect of anti-Tim-3, CD3 bi-specific antibody on co-culture of acute myeloid leukemia (AML) cells and PBMCs.

Figure 14. Chart showing effect of anti-Tim-3, CD3 bi-specific antibody on purified T cells and acute myeloid leukemia (AML) cells.

Figure 15. Chart showing effect of anti-TIM-3 clone A11-anti CD3 bispecific antibody, and anti-TIM-3 clone B10-anti CD3 bispecific antibody on purified T cells and acute myeloid leukemia (AML) cells. AML cells were mixed with purified T cells at a 1:1 ratio and the antibody was added at various concentrations. After 24-hour incubation, lysis was measured.

Figure 16. Chart showing CD34 specific cell killing effect of anti-TIM-3 clone A11-anti CD3 bispecific antibody on CD34+ cells in AML biopsies (i.e. AML stem cells). After selection, CD34+ cells (samples >99% CD34+ purity) were mixed with purified T cells at a

1:1 ratio and the antibody was added at various concentrations. After 24-hour incubation, lysis was measured.

Figure 17. Schematic drawing of the tandem single chain bispecific antibody format.

Examples

Isolation of anti-human TIM-3 antibodies

Anti-TIM-3 antibodies were isolated from a human antibody phage display library via *in vitro* selection in a 3-round bio-panning process.

Basically, streptavidin-magnetic beads were coated with biotinylated human TIM-3 and used to fish-out anti-TIM-3-specific phages using magnetic sorting. Some steps to get rid of potential anti-biotin antibodies were added in the selection process.

After a small-scale induction in HB2151 cells, Fab antibodies were screened by ELISA. Briefly, ELISA plates were coated with human Tim-3 coupled to human Fc and blocked with a solution of casein. After extensive washes in PBS Tween-20, Fab-containing supernatants from induction plates were transferred into the ELISA plates in the presence of 7% milk in PBS. After 90 minutes at room temperature under agitation and extensive washes, a goat anti-human Fab antibody coupled to HRP was added. One hour later, plates were washed and TMB substrate added. The reaction was stopped with 1M HCl and optical density measured at 450nm with a reference at 670nm. Antibodies giving an absorbance >0.1 were selected as positive. A first clonality screening was performed by DNA fingerprinting; clonality was then confirmed by sequencing.

Binding to human TIM-3 and cross-reactivity to mouse TIM-3

Binding to either human or mouse TIM-3 was assessed by ELISA as described above using either human or mouse TIM-3 coupled to human Fc as an antigen. Non-specific binding to human Fc was also assessed using human Fc as a negative control antigen (Figures 5 and 9).

Blocking the TIM-3/Galectin-9 interaction in vitro

Human TIM-3 coupled to phycoerythrin was pre-incubated for 30 minutes at room temperature with various concentrations of antibodies in FACS buffer. Such premixes

were then added on Galectin-9-expressing MOLT3 cells that were previously plated in 96-well plates and fixed/permeabilised in Fix/Perm buffer in the presence of anti-CD16/CD32 antibody. After 30 minutes of incubation at 4°C in the presence of the premixes and 3 washes in Perm/Wash buffer, cells were resuspended in PBS and analysed by flow cytometry (Figures 6 and 10).

The ability of the antibodies to block the TIM-3/Galectin-9 interaction was measured using the proportion of cells stained with phycoerythrin:

$$\frac{\text{mean MFI}_{\text{negative control}} - \text{MFI}_{\text{tested antibody}}}{\text{mean MFI}_{\text{negative control}}} \%$$

Affinity of the isolated anti-TIM-3 antibodies

Affinity of the antibodies for human TIM-3 was measured by Surface Plasmon Resonance.

Briefly, human TIM-3 coupled to human Fc was immobilised on a sensor chip compatible with the Proteon XPR36 bioanalyser (Biorad). Antibodies were then applied onto the chip as a flow. Association/dissociation rates were recorded for each candidate Fab and affinity (K_D) calculated (Figures 7 and 11).

In vitro functional activity: antibody-dependent cell toxicity (ADCC)

According to the French-American-British classification system, acute myeloid leukemia (AML) is divided in 8 different subtypes named M0 to M7, depending on the type of cells from which the disease originates and its degree of maturity. Except for M3 cells, all AML cell lines express TIM-3.

Anti-TIM-3 antibodies were tested on AML cell lines for their ability to kill TIM-3-expressing AML cells. Basically, AML cells were co-cultured with NK cells (1:1 ratio) in RPMI, 10% FBS and in the presence of 10µg/mL of antibody. After 4 hours at 37°C, cells were harvested and cell death/survival ratio was measured using calcein AM staining in a flow cytometry assay (Figures 8 and 12).

A11 expressed as IgG1 was then used to construct a bi-specific antibody able to engage T cells on one side (specificity for CD3) and target TIM-3 on the other side. The bispecific antibody comprises two single chain variable Fragments (scFvs) as a fusion protein. One of the scFvs comprises the V_H and V_L sequences for clone A11 (i.e. SEQ ID NOs: 45 and

50), and the other scFv comprises the V_H and V_L sequences for an anti-CD3 antibody clone.

The format for the tandem single chain bispecific antibodies is shown in Figure 17.

The bi-specific antibody was tested at 10µg/mL on co-culture of AML cells and PBMCs (PBMC/AML ratio 20:1) following a similar protocol (Figure 13).

The anti-TIM-3 clone A11-anti-CD3 bi-specific antibody was then tested in an assay with purified T cells and AML stem cells (1:1 ratio) following the same protocol (Figure 14).

Engineering

Clone A11 was further engineered; its sequence was reverted to a germline-like framework resulting in clone A11_gl (modifications only in the light chain variable domain).

Specific killing by bispecific Anti-TIM-3, anti-CD3 antibodies of Acute Myeloid Leukemia cells from patients Anti-TIM-3 clone A11, in tandem single-chain bispecific format with anti-CD3 (anti-TIM-3 clone A11-anti CD3 bispecific antibody), was tested to assess its ability to kill Acute Myeloid Leukamia (AML) cells obtained from AML patients' biopsies.

Briefly, purified T cells were mixed with AML cells obtained from patients refractory to 3 lines of chemotherapy treatment, at a ratio of 1:1. The bispecific antibody was added at various concentrations, and the mixture was incubated for 24 hours. After incubation, lysis of AML cells was measured.

The results are shown in Figure 15. Anti-TIM-3 clone A11-anti CD3 bispecific antibody proved to be potent in killing AML cells from chemotherapy-refractory patients *ex vivo*.

An anti-TIM-3 clone B10-anti CD3 bispecific antibody in tandem single-chain Fv format was also constructed and tested, and showed killing at high concentrations (Figure 15).

The clone A11-anti CD3 bispecific antibody was then tested on AML stem cells, i.e. cells within the AML biopsies that express high levels of CD34. After selection, CD34+ cells (samples >99% CD34+ purity) were mixed with purified T cells at a 1:1 ratio and the antibody was added at various concentrations. After 24-hour incubation, lysis was measured.

The results are shown in Figure 16. Anti-TIM-3 clone A11-anti CD3 bispecific antibody showed an ability to kill AML stem cells ex vivo.

Claims:

1. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, having the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
ii)	LC-CDR2:	AASTLQS	(SEQ ID NO:7)
iii)	LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)
iv)	HC-CDR1:	SYYMH	(SEQ ID NO:58), or
		GYTFTSYYMH	(SEQ ID NO:24)
v)	HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
vi)	HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

2. The antibody, or antigen binding fragment, of claim 1, having at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

3. The antibody, or antigen binding fragment, of claim 1 or 2, having at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	SYYMH	(SEQ ID NO:58), or
	GYTFTSYYMH	(SEQ ID NO:24)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

4. The antibody, or antigen binding fragment, of any one of claims 1 to 3, wherein the antibody is cytotoxic.

5. The antibody, or antigen binding fragment, of any one of claims 1 to 4, wherein the antibody is effective to restore T-cell function in T-cells exhibiting T-cell exhaustion or T-cell anergy.

6. An *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, according to any one of claims 1 to 5 bound to TIM-3.

7. An isolated light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

8. An isolated heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	SYYMH	(SEQ ID NO:58), or
	GYTFTSYYMH	(SEQ ID NO:24)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

9. An isolated light chain variable region polypeptide of claim 7 in combination with a heavy chain variable region polypeptide according to claim 8.

10. An antibody or antigen binding fragment which is capable of binding to TIM-3, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to SYYMH (SEQ ID NO:58) or GYTFTSYYMH (SEQ ID NO:24), IINPSGGSTSYAQKFQG (SEQ ID NO:25), SPGVVTALFDY (SEQ ID NO:26) respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQDIGSYLA (SEQ ID NO:6), AASTLQS (SEQ ID NO:7), QQSYSSPPT (SEQ ID NO:8), respectively.

11. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 19, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:1.

12. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, having the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
ii)	LC-CDR2:	DATNRAT	(SEQ ID NO:10)
iii)	LC-CDR3:	QHRRT	(SEQ ID NO:11)
iv)	HC-CDR1:	SSDYYWG	(SEQ ID NO:59), or
		GGSIGSSDYYWG	(SEQ ID NO:27)
v)	HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
vi)	HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

13. The antibody, or antigen binding fragment, of claim 12, having at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

14. The antibody, or antigen binding fragment, of claim 12 or 13, having at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	SSDYYWG	(SEQ ID NO:59), or
	GGSIGSSDYYWG	(SEQ ID NO:27)
HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

15. The antibody, or antigen binding fragment, of any one of claims 12 to 14, wherein the antibody is cytotoxic.

16. The antibody, or antigen binding fragment, of any one of claims 12 to 15, wherein the antibody is effective to restore T-cell function in T-cells exhibiting T-cell exhaustion or T-cell anergy.

17. An *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, according to any one of claims 12 to 16 bound to TIM-3.

18. An isolated light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

19. An isolated heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	SSDYYWG	(SEQ ID NO:59), or
	GGSIGSSDYYWG	(SEQ ID NO:27)
HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

20. An isolated light chain variable region polypeptide of claim 18 in combination with a heavy chain variable region polypeptide according to claim 19.

21. An antibody or antigen binding fragment which is capable of binding to TIM-3, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to SSDYYWG (SEQ ID NO:59) or GGSIGSSDYYWG (SEQ ID NO:27), SIYYSGSTYYNPSLKS (SEQ ID NO:28), GEHRGEFDY (SEQ ID NO:29), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQSVGSYLA (SEQ ID NO:9), DATNRAT (SEQ ID NO:10), QHRRT (SEQ ID NO:11), respectively.

22. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 20, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:2.

23. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, having the amino acid sequences i) to vi):

i)	LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
ii)	LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
iii)	LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)
iv)	HC-CDR1:	SSNWWS	(SEQ ID NO:60), or
		GGSISSSNWWS	(SEQ ID NO:30)
v)	HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
vi)	HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

24. The antibody, or antigen binding fragment, of claim 23, having at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

25. The antibody, or antigen binding fragment, of claim 23 or 24, having at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	SSNWWS	(SEQ ID NO:60), or
	GGSISSSNWWS	(SEQ ID NO:30)
HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

26. The antibody, or antigen binding fragment, of any one of claims 23 to 25, wherein the antibody is cytotoxic.

27. The antibody, or antigen binding fragment, of any one of claims 23 to 26, wherein the antibody is effective to restore T-cell function in T-cells exhibiting T-cell exhaustion or T-cell anergy.

28. An *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, according to any one of claims 23 to 27 bound to TIM-3.

29. An isolated light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

30. An isolated heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	SSNWWS	(SEQ ID NO:60), or
	GGSISSSNWWS	(SEQ ID NO:30)
HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

31. An isolated light chain variable region polypeptide of claim 29 in combination with a heavy chain variable region polypeptide according to claim 30.

32. An antibody or antigen binding fragment which is capable of binding to TIM-3, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to SSNWWS (SEQ ID NO:60) or GGSISSSNWWS (SEQ ID NO:30), EIYHSGSTNYNPSLKS (SEQ ID NO:31), VVAVAGTVDY (SEQ ID NO:32), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RSSQSLLHSNGYNYLD (SEQ ID NO:12), LGSNRAS (SEQ ID NO:13), MQGTHWPPT (SEQ ID NO:14),respectively.

33. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 21, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:3.

34. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, having the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
ii)	LC-CDR2:	GASSRAT	(SEQ ID NO:16)
iii)	LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)
iv)	HC-CDR1:	SYYMH	(SEQ ID NO:58), or
		GYTFTSYYMH	(SEQ ID NO:24)
v)	HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
vi)	HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

35. The antibody, or antigen binding fragment, of claim 34, having at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

36. The antibody, or antigen binding fragment, of claim 34 or 35, having at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	SYYMH	(SEQ ID NO:58), or
	GYTFTSYYMH	(SEQ ID NO:24)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

37. The antibody, or antigen binding fragment, of any one of claims 34 to 36, wherein the antibody is cytotoxic.

38. The antibody, or antigen binding fragment, of any one of claims 34 to 37, wherein the antibody is effective to restore T-cell function in T-cells exhibiting T-cell exhaustion or T-cell anergy.

39. An *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, according to any one of claims 34 to 38 bound to TIM-3.

40. An isolated light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

41. An isolated heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	SYYMH	(SEQ ID NO:58), or
	GYTFTSYYMH	(SEQ ID NO:24)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

42. An isolated light chain variable region polypeptide of claim 40 in combination with a heavy chain variable region polypeptide according to claim 41.

43. An antibody or antigen binding fragment which is capable of binding to TIM-3, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to SYYMH (SEQ ID NO:58) or GYTFTSYYMH (SEQ ID NO:24), IINPSGGSTSYAQKFQG (SEQ ID NO:25), DQYSSGWYYYGMDV (SEQ ID NO:33), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQSVSSSYLA (SEQ ID NO:15), GASSRAT (SEQ ID NO:16), QQYGSSPIT (SEQ ID NO:17), respectively.

44. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 22, and

the light chain sequence has at least 85% sequence identity to the light chain

sequence: SEQ ID NO:4.

45. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, having the amino acid sequences i) to vi):

i)	LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
ii)	LC-CDR2:	GASSRAT	(SEQ ID NO:16)
iii)	LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)
iv)	HC-CDR1:	SYYMH	(SEQ ID NO:58), or
		GYTFTSYYMH	(SEQ ID NO:24)
v)	HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
vi)	HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

46. The antibody, or antigen binding fragment, of claim 45, having at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

47. The antibody, or antigen binding fragment, of claim 45 or 46, having at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	SYYMH	(SEQ ID NO:58), or
	GYTFTSYYMH	(SEQ ID NO:24)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

48. The antibody, or antigen binding fragment, of any one of claims 45 to 47, wherein the antibody is cytotoxic.

49. The antibody, or antigen binding fragment, of any one of claims 45 to 48, wherein the antibody is effective to restore T-cell function in T-cells exhibiting T-cell exhaustion or T-cell anergy.

50. An *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, according to any one of claims 45 to 49 bound to TIM-3.

51. An isolated light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

52. An isolated heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	SYYMH	(SEQ ID NO:58), or
	GYTFTSYYMH	(SEQ ID NO:24)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

53. An isolated light chain variable region polypeptide of claim 51 in combination with a heavy chain variable region polypeptide according to claim 52.

54. An antibody or antigen binding fragment which is capable of binding to TIM-3, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to SYYMH (SEQ ID NO:58) or GYTFTSYYMH (SEQ ID NO:24), IINPSGGSTSYAQKFQG (SEQ ID NO:25), DLYSYGFYYYGMDV (SEQ ID NO:34), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to RASQSVSSSYLA (SEQ ID NO:15), GASSRAT (SEQ ID NO:16), QQYGSSPIT (SEQ ID NO:17), respectively.

55. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 23, and

the light chain sequence has at least 85% sequence identity to the light chain

sequence: SEQ ID NO:5.

56. An antibody, or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, having the amino acid sequences i) to vi):

i)	LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
ii)	LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
iii)	LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)
iv)	HC-CDR1:	GYYWS	(SEQ ID NO:61), or
		GGSFSGYYWS	(SEQ ID NO:52)
v)	HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
vi)	HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

or a variant thereof in which one or two or three amino acids in one or more of the sequences (i) to (vi) are replaced with another amino acid.

57. The antibody, or antigen binding fragment, of claim 1, having at least one light chain variable region incorporating the following CDRs:

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

58. The antibody, or antigen binding fragment, of claim 1 or 2, having at least one heavy chain variable region incorporating the following CDRs:

HC-CDR1:	GYYWS	(SEQ ID NO:61), or
	GGSFSGYYWS	(SEQ ID NO:52)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

59. The antibody, or antigen binding fragment, of any one of claims 1 to 3, wherein the antibody is cytotoxic.

60. The antibody, or antigen binding fragment, of any one of claims 1 to 4, wherein the antibody is effective to restore T-cell function in T-cells exhibiting T-cell exhaustion or T-cell anergy.

61. An *in vitro* complex, optionally isolated, comprising an antibody, or antigen binding fragment, according to any one of claims 1 to 5 bound to TIM-3.

62. An isolated light chain variable region polypeptide comprising the following CDRs:

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

63. An isolated heavy chain variable region polypeptide comprising the following CDRs:

HC-CDR1:	GYYWS	(SEQ ID NO:61), or
	GGSFSGYYWS	(SEQ ID NO:52)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

64. An isolated light chain variable region polypeptide of claim 7 in combination with a heavy chain variable region polypeptide according to claim 8.

65. An antibody or antigen binding fragment which is capable of binding to TIM-3, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain comprises a HC-CDR1, HC-CDR2, HC-CDR3, having at least 85% overall sequence identity to GYYWS (SEQ ID NO:61) or GGSFSGYYWS (SEQ ID NO:52), EINHSGSTNYNPSLKS (SEQ ID NO:53), GYVAGFDY (SEQ ID NO:54), respectively, and

the light chain comprises a LC-CDR1, LC-CDR2, LC-CDR3, having at least 85% overall sequence identity to SGSSSNIGNNYVS (SEQ ID NO:47), GNNWRPS (SEQ ID NO:48), ETWDSSLSAGV (SEQ ID NO:49), respectively.

66. An antibody or antigen binding fragment which is capable of binding to TIM-3, optionally isolated, comprising a heavy chain and a light chain variable region sequence, wherein:

the heavy chain sequence has at least 85% sequence identity to the heavy chain sequence: SEQ ID NO: 50 or 51, and

the light chain sequence has at least 85% sequence identity to the light chain sequence: SEQ ID NO:45 or 46.

67. An antibody or antigen binding fragment, optionally isolated, which is capable of binding to TIM-3, which is a bispecific antibody or a bispecific antigen binding fragment comprising (i) an antigen binding fragment or polypeptide according to any of one of claims 1 to 66, and (ii) an antigen binding domain which is capable of binding to a target protein other than TIM-3.

68. The antibody, or antigen binding fragment, of claim 67, wherein the antigen binding domain which is capable of binding to a target protein other than TIM-3 is capable of binding to CD3 or a CD3 polypeptide.

69. A composition comprising the antibody, or antigen binding fragment, or polypeptide of any one of claims 1 to 68 and at least one pharmaceutically-acceptable carrier.

70. An isolated nucleic acid encoding the antibody, or antigen binding fragment or polypeptide of any of one of claims 1 to 68.

71. A vector comprising the nucleic acid of any one of claims 70.

72. A host cell comprising the vector of claim 71.

73. A method for making an antibody, or antigen binding fragment or polypeptide of any of one of claims 1 to 68 comprising culturing the host cell of claim 72 under conditions suitable for the expression of a vector encoding the antibody, or antigen binding fragment or polypeptide, and recovering the antibody, or antigen binding fragment or polypeptide.

74. An antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 for use in therapy, or in a method of medical treatment.

75. An antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 for use in the treatment of cancer or a T-cell dysfunctional disorder.

76. An antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 for use in the treatment of cancer.

77. An antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 for use in the treatment of an infectious disease.

78. Use of an antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 in the manufacture of a medicament for use in the treatment of cancer or a T-cell dysfunctional disorder.

79. Use of an antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 in the manufacture of a medicament for use in the treatment of cancer.

80. Use of an antibody, antigen binding fragment or polypeptide according to any of one of claims 1 to 68 in the manufacture of a medicament for use in the treatment of an infectious disease.

81. A method, *in vitro* or *in vivo*, of killing a cell that expresses TIM-3 comprising administering an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 68 to a cell that expresses TIM-3.

82. A method, *in vitro* or *in vivo*, of enhancing T-cell function comprising administering an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 686 to a dysfunctional T-cell.

83. A method of treating cancer or a T-cell dysfunctional disorder comprising administering an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 68 to a patient suffering from cancer or a T-cell dysfunctional disorder.

84. A method of treating cancer comprising administering an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 68 to a patient suffering from cancer.

85. A method of treating an infectious disease comprising administering an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 68 to a patient suffering from an infectious disease.

86. A method comprising contacting a sample containing, or suspected to contain, TIM-3 with an antibody or antigen binding fragment according to any one of claims 1 to 68 and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.

87. A method of diagnosing a disease or condition in a subject, the method comprising contacting, *in vitro*, a sample from the subject with an antibody, or antigen binding fragment, according to any one of claims 1 to 68 and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.

88. A method of selecting or stratifying a subject for treatment with a modulator of TIM-3 signalling, the method comprising contacting, *in vitro*, a sample from the subject with an antibody, or antigen binding fragment, according to any one of claims 1 to 68 and detecting the formation of a complex of antibody, or antigen binding fragment, and TIM-3.

89. Use of an antibody, or antigen binding fragment, according to any one of claims 1 to 68 for the detection of TIM-3 *in vitro*.

90. Use of an antibody, or antigen binding fragment, according to any one of claims 1 to 68 as an *in vitro* diagnostic agent.

91. A method for expanding a population of T cells, wherein T cells are contacted *in vitro* or *ex vivo* with an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 68.

92. A method of treatment of a subject having a T-cell dysfunctional disorder, the method comprising culturing T cells obtained from a blood sample from a subject in the presence of an antibody, antigen binding fragment or polypeptide according to any one of claims 1 to 68 so as to expand the T cell population, collecting expanded T cells, and administering the expanded T cells to a subject in need of treatment.

A3 clone

DIQMTQSPSFMSASVGDRVTITCRASQDIGSYLAWYQQKPGKAPKLLIYAASTL
QSGVPSRFSGSGSGTDFTLTINSLQPEDFATYYCQQSYSSPPTFGPGTTLEIK
(SEQ ID NO:1)

LC-CDR1:	RASQDIGSYLA	(SEQ ID NO:6)
LC-CDR2:	AASTLQS	(SEQ ID NO:7)
LC-CDR3:	QQSYSSPPT	(SEQ ID NO:8)

B10 clone

EIVLTQSPATLSFSPGERATLSCRASQSVGSYLAWYQQRPGQAPRPLIYDATN
RATGIPTRFSGSGSGTDFTLTISSLEPEDFATYYCQHRRTFGRGTKLEIK
(SEQ ID NO:2)

LC-CDR1:	RASQSVGSYLA	(SEQ ID NO:9)
LC-CDR2:	DATNRAT	(SEQ ID NO:10)
LC-CDR3:	QHRRT	(SEQ ID NO:11)

G6 clone

DVVMTQSPLSLPVTPGEPASISCRSSQSLLHSNGYNYLDWYLQKPGQSPQLLI
YLGSNRASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQGTHWPPTFGQ
GTKVELK (SEQ ID NO:3)

LC-CDR1:	RSSQSLLHSNGYNYLD	(SEQ ID NO:12)
LC-CDR2:	LGSNRAS	(SEQ ID NO:13)
LC-CDR3:	MQGTHWPPT	(SEQ ID NO:14)

Figure 1

G7 clone

ETTLTQSPGTLSLSPGERATLSCRASQSVSSSYLAWYQQTPGQAPRLLIYGAS
SRATGIPDRFSGSGSGTDFTLTISRLEPEDFAVYYCQQYGSSPITFGQGTRLEIK
(SEQ ID NO:4)

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

G9 clone

ETTLTQSPGTLSLSPGERATLSCRASQSVSSSYLAWYQQKIGQAPRLLIYGASS
RATGIPDRFSGSGSGTDFTLTISRLEPEDFAVYYCQQYGSSPITFGQGTRLEIK
(SEQ ID NO:5)

LC-CDR1:	RASQSVSSSYLA	(SEQ ID NO:15)
LC-CDR2:	GASSRAT	(SEQ ID NO:16)
LC-CDR3:	QQYGSSPIT	(SEQ ID NO:17)

A11 clone

QSVVTQPPSVSAAPGQKVTISCSGSSSNIGNNYVSWYQQLPGTAPKLLIYGNN
WRPSGIPDRFSGSKSGTSATLAISGLQTGDEADYYCETWDSSLSAGVFGGGT
KLTVL (SEQ ID NO:45)

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

Figure 1 (Cont.)

A11_gl clone

QSVLTQPPSVSAAPGQKVTISCSGSSSNIGNNYVSWYQQLPGTAPKLLIYGNN
WRPSGIPDRFSGSKSGTSATLGITGLQTGDEADYYCETWDSSLSAGVFGGGT
KLTVL (SEQ ID NO:46)

LC-CDR1:	SGSSSNIGNNYVS	(SEQ ID NO:47)
LC-CDR2:	GNNWRPS	(SEQ ID NO:48)
LC-CDR3:	ETWDSSLSAGV	(SEQ ID NO:49)

Figure 1 (Cont.)

A3 clone

QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGII
NPSGGSTSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARSPGVV
TALFDYWGQGTLVTVSS (SEQ ID NO:19)

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	SPGVVTALFDY	(SEQ ID NO:26)

B10 clone

QVQLQESGPGLVKASETLSLTCTVSGGSIGSSDYYWGWIRQPPGKGLEWIGSI
YYSGSTYYNPSLKSRVTMSVDTPNNQFSLKLSSVTAADTAVYYCARGEHRGEF
DYWGQGTLVTVSS (SEQ ID NO:20)

HC-CDR1:	GGSIGSSDYYWG	(SEQ ID NO:27), or
	SSDYYWG	(SEQ ID NO:59)
HC-CDR2:	SIYYSGSTYYNPSLKS	(SEQ ID NO:28)
HC-CDR3:	GEHRGEFDY	(SEQ ID NO:29)

G6 clone

QVQLQESGPGLVKPSGTLSLTCAVSGGSISSSNWWSWVRQPPGKGLEWIGEI
YHSGSTNYNPSLKSRVTISVDKSKNQFSLKLSSVTAADTAVYYCARVVAVAGTV
DYWGQGTLVTVSS (SEQ ID NO:21)

HC-CDR1:	GGSISSSNWWS	(SEQ ID NO:30), or
	SSNWWS	(SEQ ID NO:60)
HC-CDR2:	EIYHSGSTNYNPSLKS	(SEQ ID NO:31)
HC-CDR3:	VVAVAGTVDY	(SEQ ID NO:32)

Figure 2

G7 clone

EVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIIN PSGGSTSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARDQYSSG WYYYGMDVWGQGTTVTVSS (SEQ ID NO:22)

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DQYSSGWYYYGMDV	(SEQ ID NO:33)

G9 clone

QVQLQQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGII NPSGGSTSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARDLYSY GFYYYGMDVWGQGTTVTVSS (SEQ ID NO:23)

HC-CDR1:	GYTFTSYYMH	(SEQ ID NO:24), or
	SYYMH	(SEQ ID NO:58)
HC-CDR2:	IINPSGGSTSYAQKFQG	(SEQ ID NO:25)
HC-CDR3:	DLYSYGFYYYGMDV	(SEQ ID NO:34)

A11 clone

QVQLQQWGAGLLKPSETLSLTCAVYGGSFSGYYWSWIRQPPGKGLEWIGEIN HSGSTNYNPSLKSRVTISVDTSKNQFSLKLSSVTAADTAVYYCARGYVAGFDY WGQGTLVTVSS (SEQ ID NO:50)

HC-CDR1:	GGSFSGYYWS	(SEQ ID NO:52), or
	GYYWS	(SEQ ID NO: 61)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

Figure 2 (Cont.)

A11 gl clone

QVQLQQWGAGLLKPSETLSLTCAVYGGSFSGYYWSWIRQPPGKGLEWIGEIN
HSGSTNYNPSLKSRVTISVDTSKNQFSLKLSSVTAADTAVYYCARGYVAGFDY
WGQGTLVTVSS (SEQ ID NO:51)

HC-CDR1:	GGSFSGYYWS	(SEQ ID NO:52), or
	GYYWS	(SEQ ID NO: 61)
HC-CDR2:	EINHSGSTNYNPSLKS	(SEQ ID NO:53)
HC-CDR3:	GYVAGFDY	(SEQ ID NO:54)

Figure 2 (Cont.)

Clone	CDR L1	CDR L2	CDR L3
Light Chain			
A3	RASQDIGSYLA (SEQ ID NO:6)	AASTLQS (SEQ ID NO:7)	QQSYSSPPT (SEQ ID NO:8)
B10	RASQSVGSYLA (SEQ ID NO:9)	DATNRAT (SEQ ID NO:10)	QHRRT (SEQ ID NO:11)
G6	RSSQSLLHSNGYNYLD (SEQ ID NO:12)	LGSNRAS (SEQ ID NO:13)	MQGTHWPPT (SEQ ID NO:14)
G7	RASQSVSSSYLA (SEQ ID NO:15)	GASSRAT (SEQ ID NO:16)	QQYGSSPIT (SEQ ID NO:17)
G9	RASQSVSSSYLA (SEQ ID NO:15)	GASSRAT (SEQ ID NO:16)	QQYGSSPIT (SEQ ID NO:17)
A11	SGSSSNIGNNYVS (SEQ ID NO:47)	GNNWRPS (SEQ ID NO:48)	ETWDSSLSAGV (SEQ ID NO:49)
A11_gl	SGSSSNIGNNYVS (SEQ ID NO:47)	GNNWRPS (SEQ ID NO:48)	ETWDSSLSAGV (SEQ ID NO:49)

Figure 3

Clone	CDR L1	CDR L2	CDR L3
		Heavy Chain	
A3	GYTFTSYYMH (SEQ ID NO:24), or SYYMH (SEQ ID NO:58)	IINPSGGSTSYAQKFQG (SEQ ID NO:25)	SPGVVTALFDY (SEQ ID NO:26)
B10	GGSIGSSDYYWG (SEQ ID NO:27), or SSDYYWG (SEQ ID NO: 59)	SIYYSGSTYYNPSLKS (SEQ ID NO:28)	GEHRGEFDY (SEQ ID NO:29)
G6	GGSISSSNWWS (SEQ ID NO:30), or SSNWWS (SEQ ID NO:60)	EIYHSGSTNYNPSLKS (SEQ ID NO:31)	VVAVAGTVDY (SEQ ID NO:32)
G7	GYTFTSYYMH (SEQ ID NO:24), or SYYMH (SEQ ID NO:58)	IINPSGGSTSYAQKFQG (SEQ ID NO:25)	DQYSSGWYYYGMDV (SEQ ID NO:33)
G9	GYTFTSYYMH (SEQ ID NO:24), or SYYMH (SEQ ID NO:58)	IINPSGGSTSYAQKFQG (SEQ ID NO:25)	DLYSYGFYYYGMDV (SEQ ID NO:34)
A11	GGSFSGYYWS (SEQ ID NO:52), or SGYYWS (SEQ ID NO:61)	EINHSGSTNYNPSLKS (SEQ ID NO:53)	GYVAGFDY (SEQ ID NO:54)
A11_gl	GGSFSGYYWS (SEQ ID NO:52), or SGYYWS (SEQ ID NO:61)	EINHSGSTNYNPSLKS (SEQ ID NO:53)	GYVAGFDY (SEQ ID NO:54)

Figure 3 (Cont.)

Light chain variable domains

A3 clone

>A3_aa_L

DIQMTQSPSFMSASVGDRVTITCRASQDIGSYLAWYQQKPGKAPKLLIYAASTLQSGVP
SRFSGSGSGTDFTLTINSLQPEDFATYYCQQSYSSPPTFGPGTTLEIK [SEQ ID
NO. 1]

>A3_ntd_L

GACATCCAGATGACCCAGTCTCCCTCCTTCATGTCTGCATCTGTAGGAGACAGAGTCAC
CATCACTTGCCGGGCCAGTCAGGACATTGGCAGTTATTTAGCCTGGTATCAGCAAAAAC
CAGGGAAAGCCCCTAAACTCCTGATCTATGCTGCATCCACTTTGCAAAGTGGGGTCCCA
TCAAGGTTCAGTGGCAGTGGATCTGGGACAGATTTCACTCTCACCATCAACAGTCTGCA
ACCTGAAGATTTTGCAACTTACTACTGTCAACAGAGTTACAGTTCCCCTCCGACTTTCG
GCCCTGGGACCACATTGGAGATCAAA [SEQ ID NO. 35]

B10 clone

>B10_aa_L

EIVLTQSPATLSFSPGERATLSCRASQSVGSYLAWYQQRPGQAPRPLIYDATNRATGIP
TRFSGSGSGTDFTLTISSLEPEDFATYYCQHRRTFGRGTKLEIK [SEQ ID NO. 2]

>B10_ntd_L

GAAATTGTGCTGACTCAGTCTCCAGCCACCCTGTCTTTTTCTCCGGGTGAAAGAGCCAC
CCTCTCCTGCAGGGCCAGTCAGAGTGTTGGCAGCTACTTAGCCTGGTACCAGCAGAGAC
CTGGCCAGGCTCCCAGGCCCCTCATCTATGATGCAACCAACAGGGCCACTGGCATCCCA
ACCAGGTTCAGTGGCAGTGGGTCTGGGACAGACTTCACTCTCACCATCAGCAGCCTAGA
GCCTGAAGATTTTGCAACTTATTACTGTCAACACCGAAGGACTTTTGGCCGGGGGACCA
AGTTGGAGATCAAA [SEQ ID NO. 36]

Figure 4

G6 clone

>G6_aa_L

DVVMTQSPLSLPVTPGEPASISCRSSQSLLHSNGYNYLDWYLQKPGQSPQLLIYLGSNR
ASGVPDRFSGSGSGTDFTLKISRVEAEDVGVYYCMQGTHWPPTFGQGTKVELK [SEQ
ID NO. 3]

>G6_ntd_L

GATGTTGTGATGACTCAGTCTCCACTCTCCCTGCCCGTCACCCCTGGAGAGCCGGCCTC
CATCTCCTGCAGGTCTAGTCAGAGCCTCCTACATAGTAATGGATACAACTATTTGGATT
GGTACCTGCAGAAGCCAGGGCAGTCTCCACAGCTCCTGATCTATTTGGGTTCTAATCGG
GCCTCCGGGGTCCCTGACAGGTTCAGTGGCAGTGGATCAGGCACAGATTTTACACTGAA
AATCAGCAGGGTGGAGGCTGAGGATGTTGGGGTTTATTACTGCATGCAAGGTACACACT
GGCCTCCGACGTTCGGCCAAGGGACCAAGGTGGAACTCAAA [SEQ ID NO. 37]

G7 clone

>G7_aa_L

ETTLTQSPGTLSLSPGERATLSCRASQSVSSSYLAWYQQTPGQAPRLLIYGASSRATGI
PDRFSGSGSGTDFTLTISRLEPEDFAVYYCQQYGSSPITFGQGTRLEIK [SEQ ID
NO. 4]

>G7_ntd_L

GAAACGACACTCACGCAGTCTCCAGGCACCCTGTCTTTGTCTCCAGGGGAAAGAGCCAC
CCTCTCCTGCAGGGCCAGTCAGAGTGTTAGCAGCAGCTACTTAGCCTGGTACCAGCAGA
CACCTGGCCAGGCTCCCAGGCTCCTCATCTATGGTGCATCCAGCAGGGCCACTGGCATC
CCAGACAGGTTCAGTGGCAGTGGGTCTGGGACAGACTTCACTCTCACCATCAGCAGACT
GGAGCCTGAAGATTTTGCAGTGTATTACTGTCAGCAGTATGGTAGCTCACCCATCACCT
TCGGCCAAGGGACACGACTGGAGATTAAA [SEQ ID NO. 38]

Figure 4 (Cont.)

G9 clone

>G9_aa_L

ETTLTQSPGTLSLSPGERATLSCRASQSVSSSYLAWYQQKIGQAPRLLIYGASSRATGI
PDRFSGSGSGTDFTLTISRLEPEDFAVYYCQQYGSSPITFGQGTRLEIK [SEQ ID
NO. 5]

>G9_ntd_L

GAAACGACACTCACGCAGTCTCCAGGCACCCTGTCTTTGTCTCCAGGGGAAAGAGCCAC
CCTCTCCTGCAGGGCCAGTCAGAGTGTTAGCAGCAGCTACTTAGCCTGGTACCAGCAAA
AAATTGGCCAGGCTCCCAGGCTCCTCATCTATGGTGCATCCAGCAGGGCCACTGGCATC
CCAGACAGGTTCAGTGGCAGTGGGTCTGGGACAGACTTCACTCTCACCATCAGCAGACT
GGAGCCTGAAGATTTTGCAGTGTATTACTGTCAGCAGTATGGTAGCTCACCAATCACCT
TCGGCCAAGGGACACGACTGGAGATTAAA [SEQ ID NO. 39]

A11 clone

>A11_aa_L

QSVVTQPPSVSAAPGQKVTISCSGSSSNIGNNYVSWYQQLPGTAPKLLIYGNNWRPSGI
PDRFSGSKSGTSATLAISGLQTGDEADYYCETWDSSLSAGVFGGGTKLTVL [SEQ ID
NO. 45]

>A11_ntd_L

CAGTCTGTCGTGACGCAGCCGCCCTCAGTGTCTGCGGCCCCAGGACAGAAAGTCACCAT
CTCCTGCTCTGGAAGCAGCTCCAACATTGGGAATAATTATGTATCCTGGTACCAGCAGC
TCCCAGGAACAGCCCCCAAACTCCTCATTTATGGCAATAATTGGCGACCCTCAGGGATT
CCTGACCGCTTCTCTGGCTCCAAGTCTGGCACCTCAGCCACCCTGGCCATCAGCGGACT
TCAGACTGGGGACGAGGCCGATTATTACTGCGAAACATGGGATAGCAGCCTGAGTGCTG
GGGTATTCGGCGGAGGGACCAAGCTGACCGTCCTA [SEQ ID NO. 55]

Figure 4 (Cont.)

A11_gl clone

>A11_gl_aa_L

QSVLTQPPSVSAAPGQKVTISCSGSSSNIGNNYVSWYQQLPGTAPKLLIYGNNWRPSGI
PDRFSGSKSGTSATLGITGLQTGDEADYYCETWDSSLSAGVFGGGTKLTVL [SEQ ID
NO. 46]

>A11_gl_ntd_L

CAGTCTGTGTTGACGCAGCCGCCCTCAGTGTCTGCGGCCCCAGGACAGAAAGTCACCAT
CTCCTGCTCTGGAAGCAGCTCCAACATTGGGAATAATTATGTATCCTGGTACCAGCAGC
TCCCAGGAACAGCCCCCAAACTCCTCATTTATGGCAATAATTGGCGACCCTCAGGGATT
CCTGACCGCTTCTCTGGCTCCAAGTCTGGCACCTCAGCCACCCTGGGCATCACCGGACT
TCAGACTGGGGACGAGGCCGATTATTACTGCGAAACATGGGATAGCAGCCTGAGTGCTG
GGGTATTCGGCGGAGGGACCAAGCTGACCGTCCTA [SEQ ID NO. 56]

Figure 4 (Cont.)

Heavy chain variable domains

A3 clone

>A3_aa_H

QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGSTS
YAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARSPGVVTALFDYWGQGTLVTV
SS [SEQ ID NO. 19]

>A3_ntd_H

CAGGTGCAGCTGGTGCAGTCTGGGGCTGAGGTGAAGAAGCCTGGGGCCTCAGTGAAGGT
TTCCTGCAAGGCATCTGGATACACCTTCACCAGCTACTATATGCACTGGGTGCGACAGG
CCCCTGGACAAGGGCTTGAGTGGATGGGAATAATCAACCCTAGTGGTGGTAGCACAAGC
TACGCACAGAAGTTCCAGGGCAGAGTCACCATGACCAGGGACACGTCCACGAGCACAGT
CTACATGGAGCTGAGCAGCCTGAGATCTGAGGACACGGCCGTGTATTACTGTGCGAGAA
GCCCTGGGGTGGTGACTGCCCTCTTTGACTACTGGGGCCAGGGAACCCTGGTCACCGTC
TCAAGC [SEQ ID NO. 40]

B10 clone

>B10_aa_H

QVQLQESGPGLVKASETLSLTCTVSGGSIGSSDYYWGWIRQPPGKGLEWIGSIYYSGST
YYNPSLKSRVTMSVDTPNNQFSLKLSSVTAADTAVYYCARGEHRGEFDYWGQGTLVTVS
S [SEQ ID NO. 20]

>B10_ntd_H

CAGGTGCAGCTGCAGGAGTCGGGCCCAGGACTGGTGAAGGCTTCGGAGACCCTGTCCCT
CACCTGCACTGTCTCTGGTGGCTCCATCGGCAGTAGTGATTACTACTGGGGCTGGATCC
GCCAGCCCCCAGGGAAGGGACTGGAGTGGATTGGGAGTATCTATTATAGTGGGAGCACC
TACTACAACCCCTCCCTCAAGAGTCGAGTCACCATGTCAGTAGACACGCCCAACAATCA
GTTCTCCCTGAAGCTGAGCTCTGTGACCGCTGCGGACACGGCCGTGTATTACTGTGCGA
GAGGAGAACATAGAGGGGAATTTGACTACTGGGGCCAGGGCACCCTGGTCACCGTCTCA
AGC [SEQ ID NO. 41]

Figure 4 (Cont.)

G6 clone

>G6_aa_H

QVQLQESGPGLVKPSGTLSLTCAVSGGSISSSNWWSWVRQPPGKGLEWIGEIYHSGSTN
YNPSLKSRVTISVDKSKNQFSLKLSSVTAADTAVYYCARVVAVAGTVDYWGQGTLVTVS
S [SEQ ID NO. 21]

>G6_ntd_H

CAGGTGCAGCTGCAGGAGTCCGGCCCAGGACTGGTGAAGCCTTCGGGGACCCTGTCCCT
CACCTGCGCTGTCTCTGGTGGCTCCATTAGCAGTAGTAACTGGTGGAGTTGGGTCCGCC
AGCCCCCAGGGAAGGGGCTGGAGTGGATTGGGGAAATCTATCATAGTGGGAGCACCAAC
TACAACCCGTCCCTCAAGAGTCGAGTCACCATATCAGTAGACAAGTCCAAGAACCAGTT
CTCCCTGAAGCTGAGCTCTGTGACCGCCGCGGACACGGCCGTGTATTACTGTGCGAGAG
TAGTAGCAGTGGCTGGTACGGTTGACTACTGGGGCCAGGGAACCCTGGTCACCGTCTCA
AGC [SEQ ID NO. 42]

G7 clone

>G7_aa_H

EVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGSTS
YAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARDQYSSGWYYYGMDVWGQGTT
VTVSS [SEQ ID NO. 22]

>G7_ntd_H

GAGGTCCAGCTGGTGCAGTCTGGGGCTGAGGTGAAGAAGCCTGGGGCCTCAGTGAAGGT
TTCCTGCAAGGCATCTGGATACACCTTCACCAGCTACTATATGCACTGGGTGCGACAGG
CCCCTGGACAAGGGCTTGAGTGGATGGGAATAATCAACCCTAGTGGTGGTAGCACAAGC
TACGCACAGAAGTTCCAGGGCAGAGTCACCATGACCAGGGACACGTCCACGAGCACAGT
CTACATGGAGCTGAGCAGCCTGAGATCTGAGGACACGGCCGTGTATTACTGTGCGAGAG
ATCAGTATAGCAGTGGCTGGTACTACTACGGTATGGACGTCTGGGGCCAAGGGACCACG
GTCACCGTCTCAAGC [SEQ ID NO. 43]

Figure 4 (Cont.)

G9 clone

>G9_aa_H

QVQLQQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGSTS
YAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARDLYSYGFYYYGMDVWGQGTT
VTVSS [SEQ ID NO. 23]

>G9_ntd_H

CAGGTACAGCTGCAGCAGTCAGGGGCTGAGGTGAAGAAGCCTGGGGCCTCAGTGAAGGT
TTCCTGCAAGGCATCTGGATACACCTTCACCAGCTACTATATGCACTGGGTGCGACAGG
CCCCTGGACAAGGGCTTGAGTGGATGGGAATAATCAACCCTAGTGGTGGTAGCACAAGC
TACGCACAGAAGTTCCAGGGCAGAGTCACCATGACCAGGGACACGTCCACGAGCACAGT
CTACATGGAGCTGAGCAGCCTGAGATCTGAGGACACGGCCGTGTATTACTGTGCGAGAG
ATTTATACAGCTATGGTTTTTACTACTACGGTATGGACGTCTGGGGCCAAGGGACCACG
GTCACCGTCTCAAGC [SEQ ID NO. 44]

A11 clone

>A11_aa_H

QVQLQQWGAGLLKPSETLSLTCAVYGGSFSGYYWSWIRQPPGKGLEWIGEINHSGSTNY
NPSLKSRVTISVDTSKNQFSLKLSSVTAADTAVYYCARGYVAGFDYWGQGTLVTVSS
[SEQ ID NO. 50]

>A11_ntd_H

CAGGTGCAGCTACAGCAGTGGGGCGCAGGACTGTTGAAGCCTTCGGAGACCCTGTCCCT
CACCTGCGCTGTCTATGGTGGGTCCTTCAGTGGTTACTACTGGAGCTGGATCCGCCAGC
CCCCAGGGAAGGGGCTGGAGTGGATTGGGGAAATCAATCATAGTGGAAGCACCAACTAC
AACCCGTCCCTCAAGAGTCGAGTCACCATATCAGTAGACACGTCCAAGAACCAGTTCTC
CTTGAAACTGAGCTCTGTGACCGCCGCGGACACGGCTGTGTATTACTGTGCGAGAGGCT
ACGTGGCTGGCTTTGACTACTGGGGCCAGGGAACCCTGGTCACCGTCTCAAGC [SEQ
ID NO. 57]

A11_gl clone heavy chain is identical to A11

Figure 4 (Cont.)

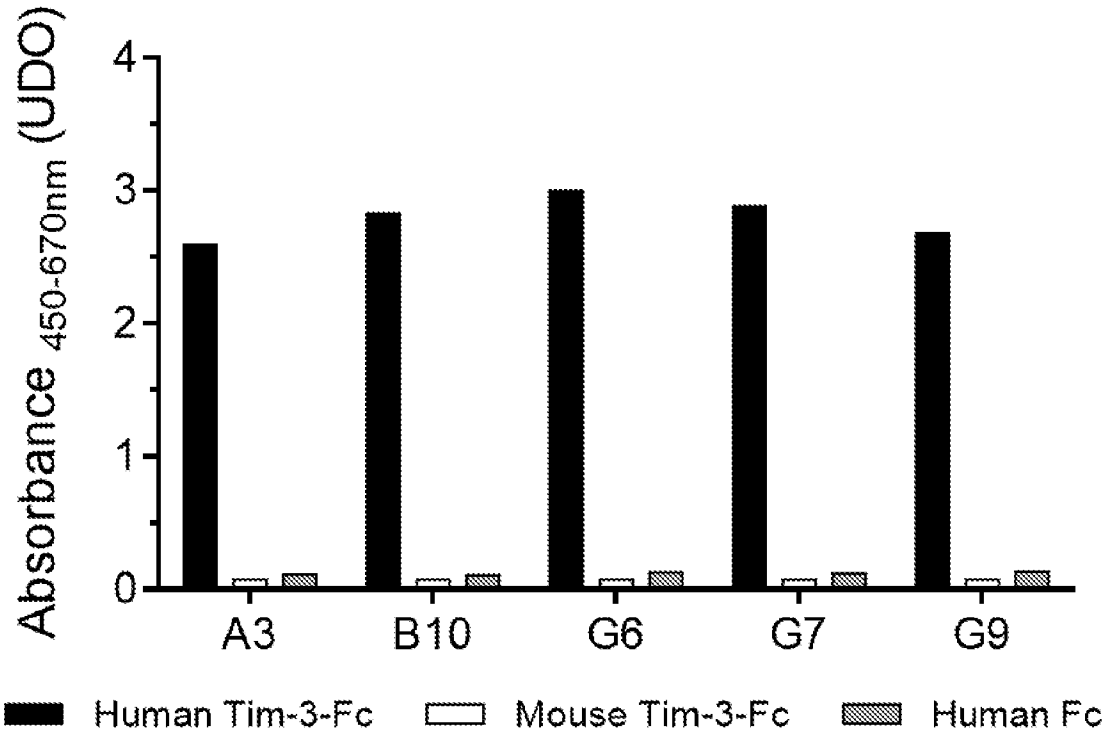

Absorbance 450-670nm (UDO)
4
3
2
1
0
A3
B10
G6
G7
G9
Human Tim-3-Fc
Mouse Tim-3-Fc
Human Fc

Figure 5

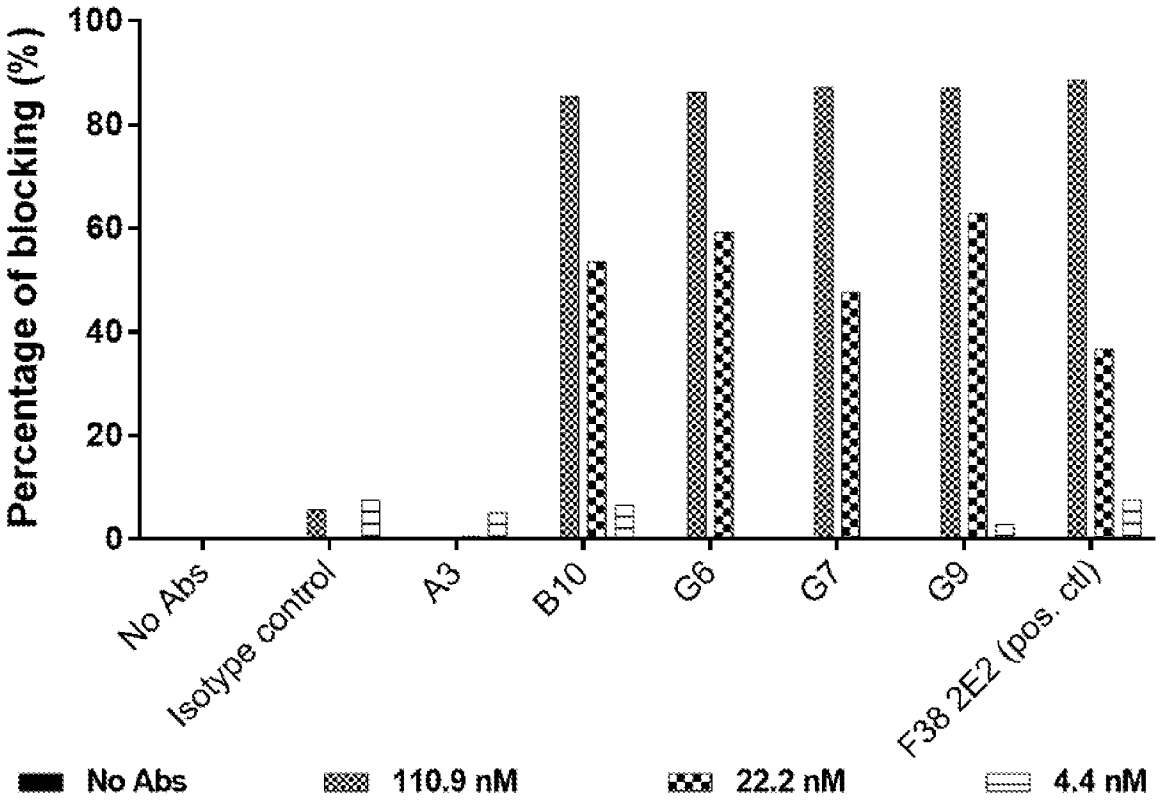

Percentage of blocking (%)
100
80
60
40
20
0
No Abs
Isotype control
A3
B10
G6
G7
G9
F38 2E2 (pos. ctl)
No Abs
110.9 nM
22.2 nM
4.4 nM

Figure 6

	Affinity (K_D in nM)
A3	0.44
B10	0.26
G6	2.52
G7	0.62
G9	0.63

Figure 7

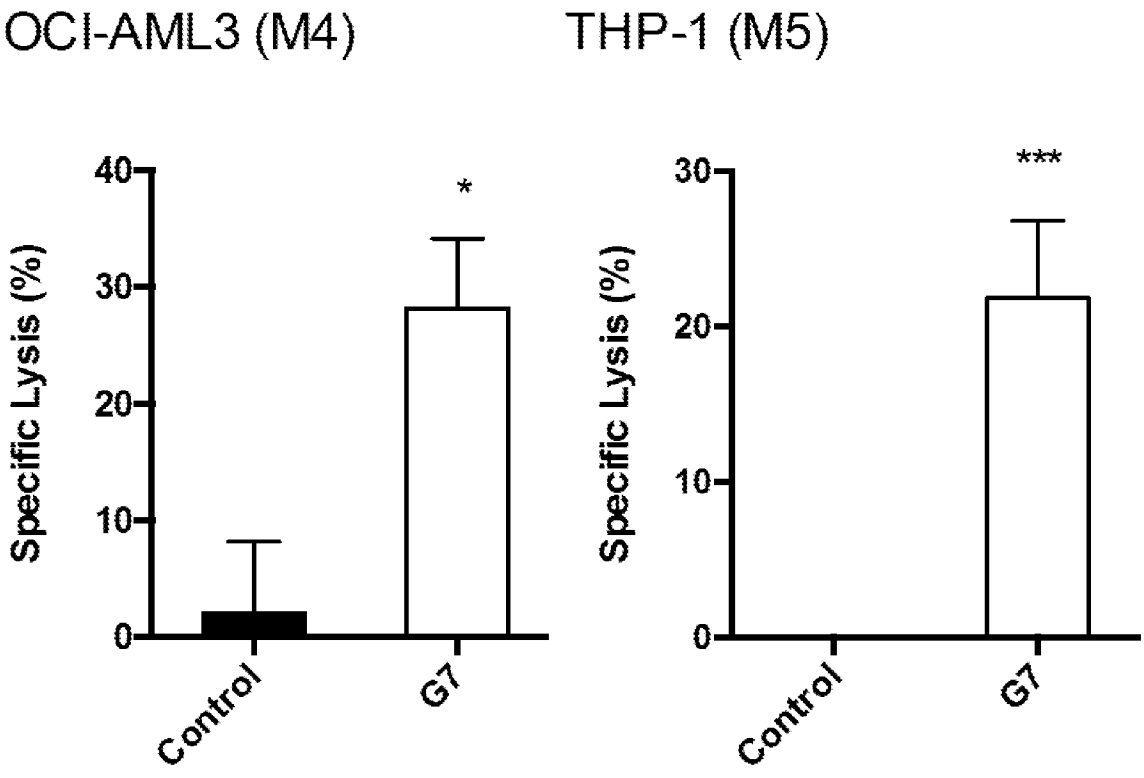


Figure 8

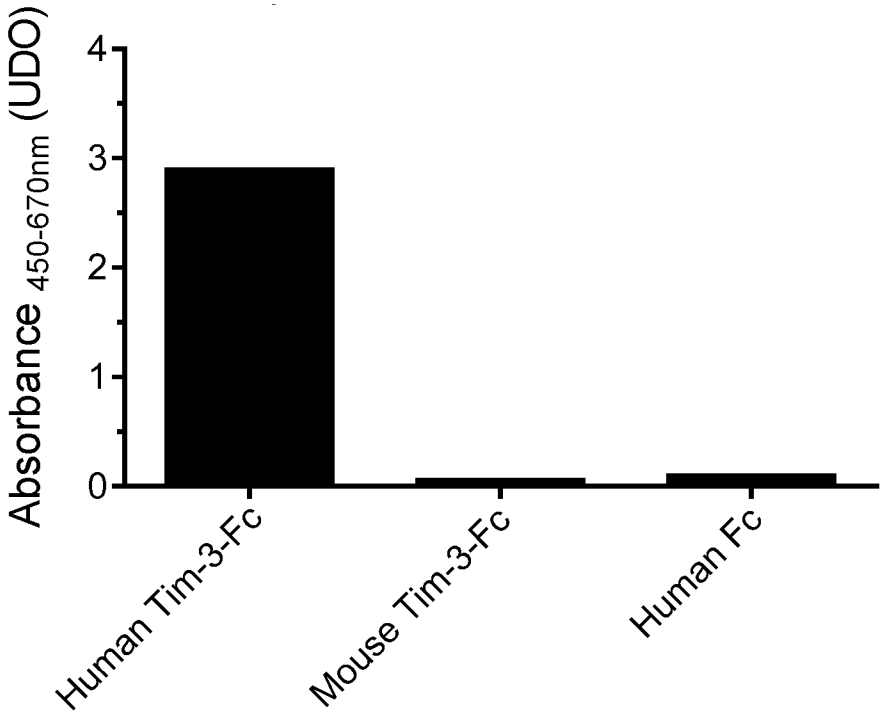


Figure 9

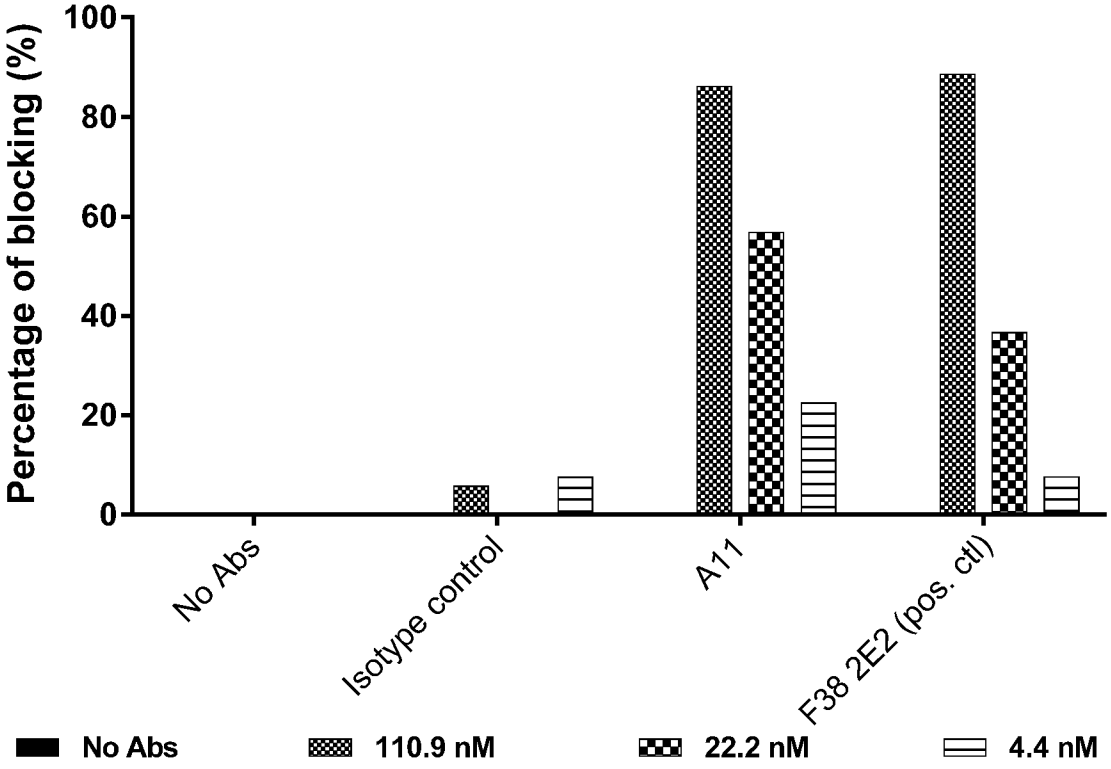


Figure 10

	Affinity (K_D in nM)
A11	1.26

Figure 11

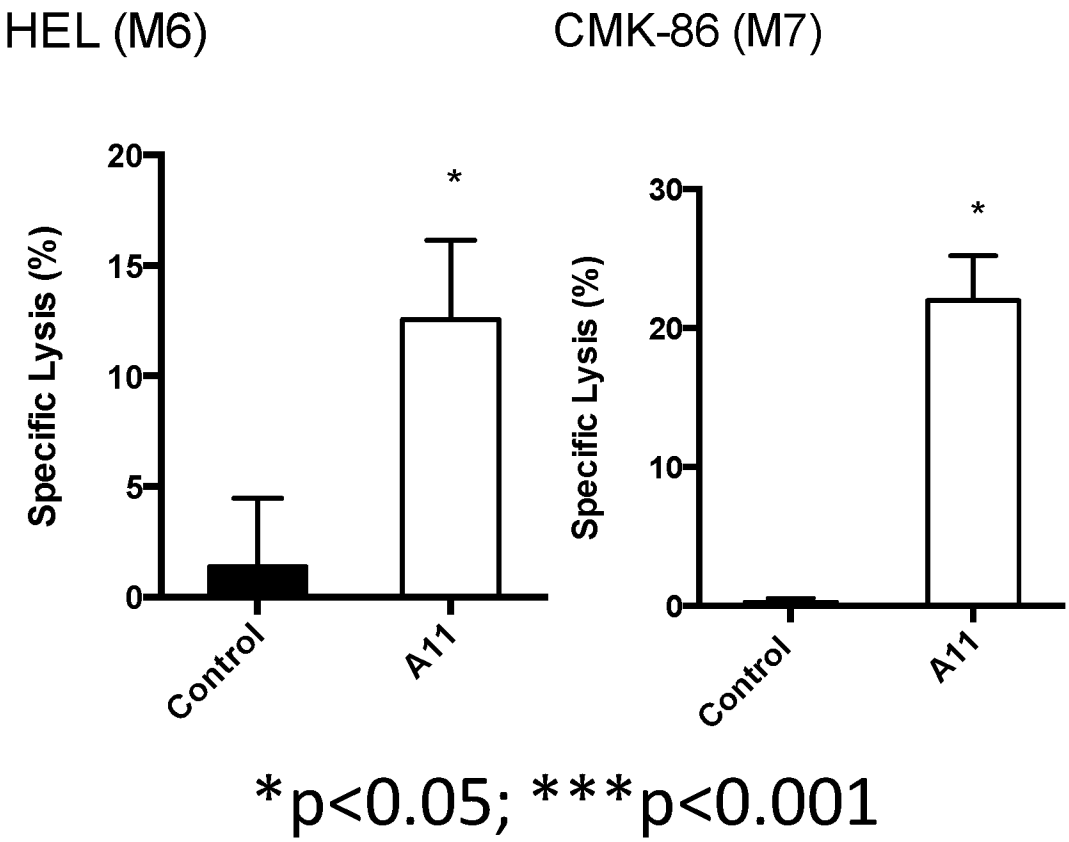


Figure 12

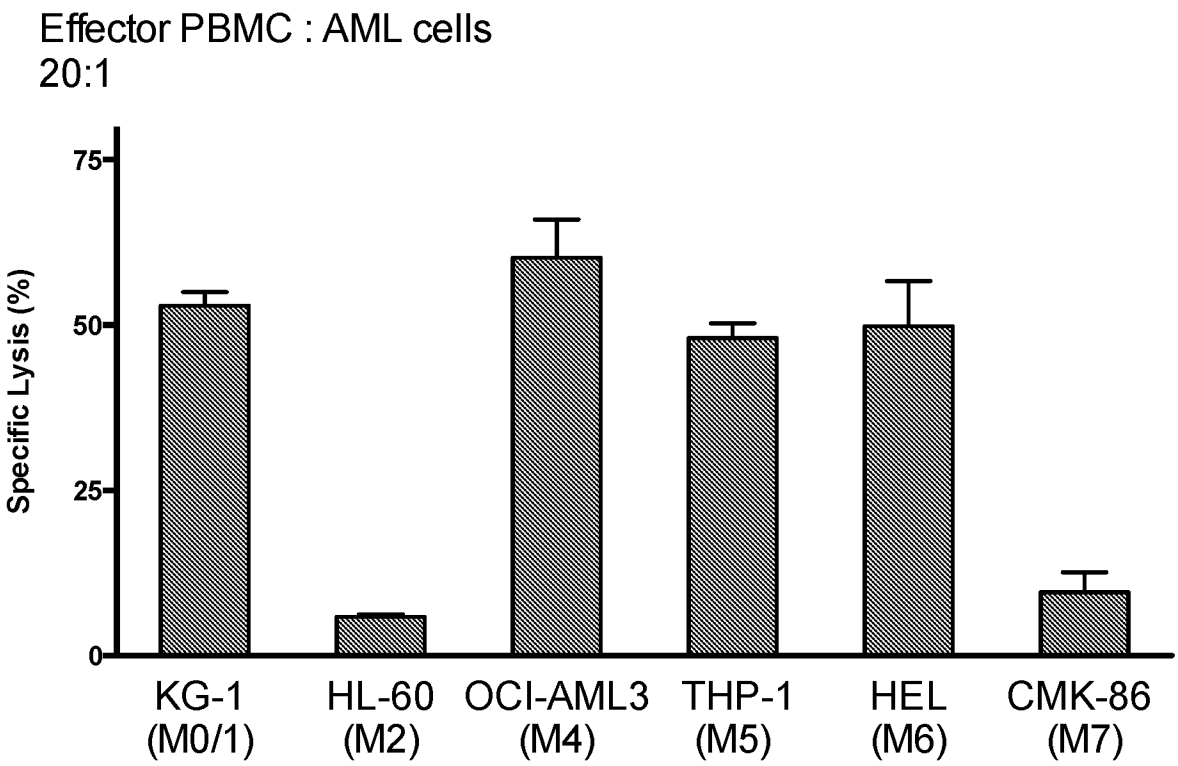

Effector PBMC : AML cells
20:1
Specific Lysis (%)
75
50
25
0
KG-1
(M0/1)
HL-60
(M2)
OCI-AML3
(M4)
THP-1
(M5)
HEL
(M6)
CMK-86
(M7)

Figure 13

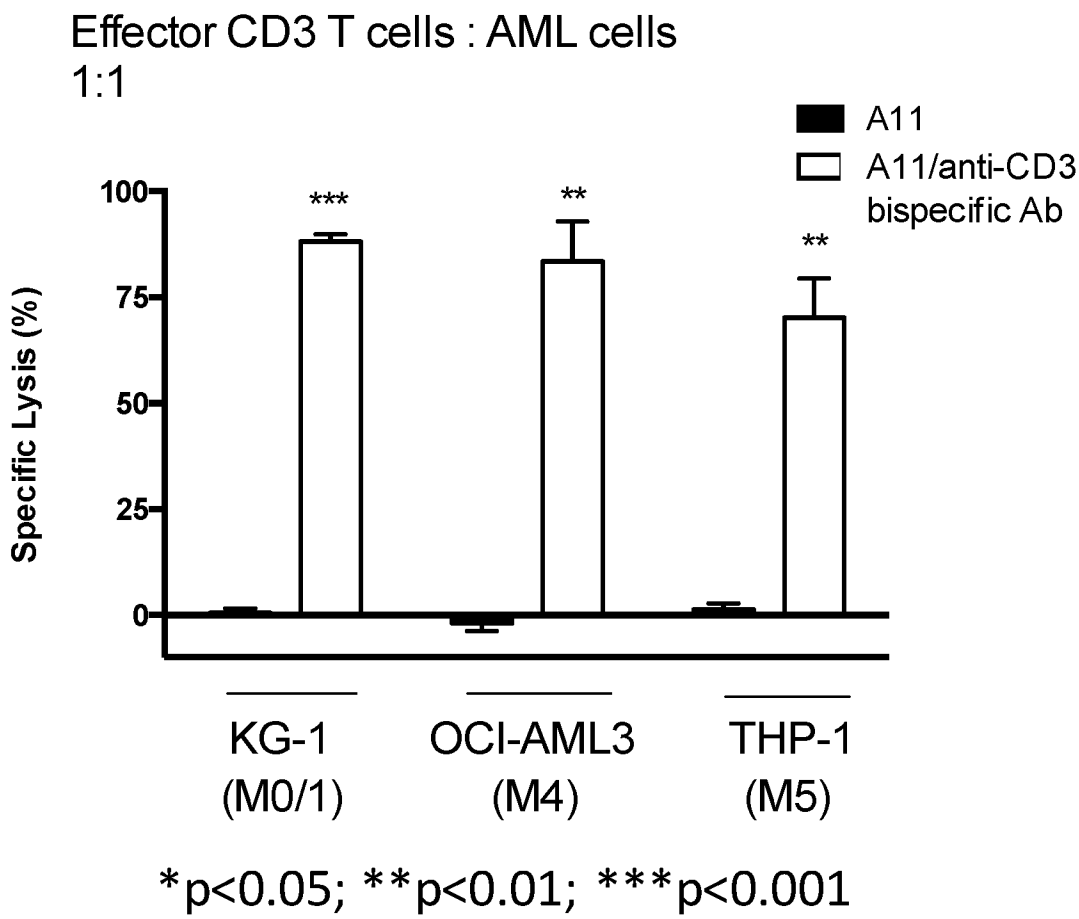

Effector CD3 T cells : AML cells
1:1
A11
A11/anti-CD3
bispecific Ab
Specific Lysis (%)
100
75
50
25
0
***
**
**
KG-1
(M0/1)
OCI-AML3
(M4)
THP-1
(M5)
*p<0.05; **p<0.01; ***p<0.001

Figure 14

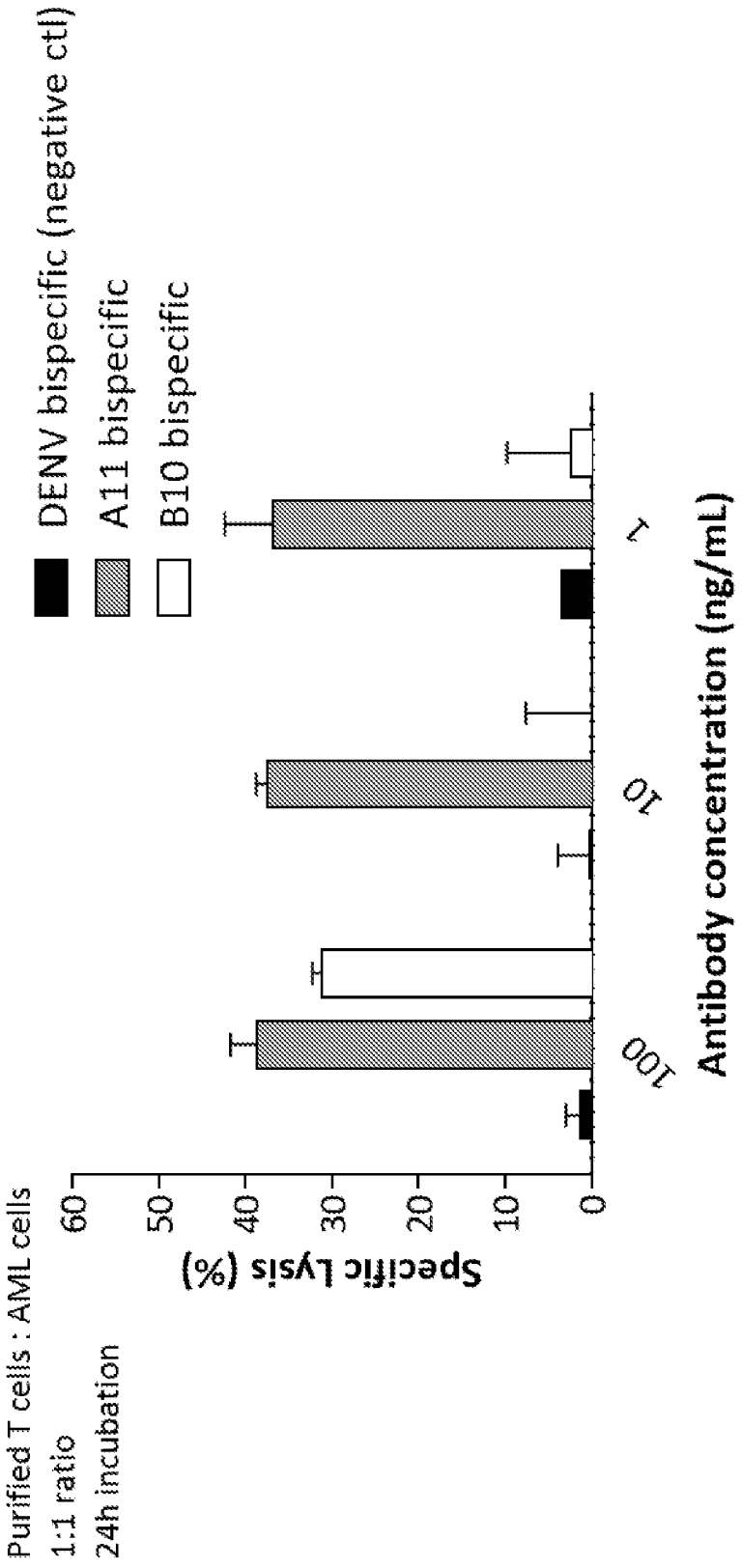

DENV bispecific (negative ctl)
A11 bispecific
B10 bispecific
Purified T cells : AML cells
1:1 ratio
24h incubation
Specific Lysis (%)
0
10
20
30
40
50
60
100
10
1
Antibody concentration (ng/mL)

Figure 15

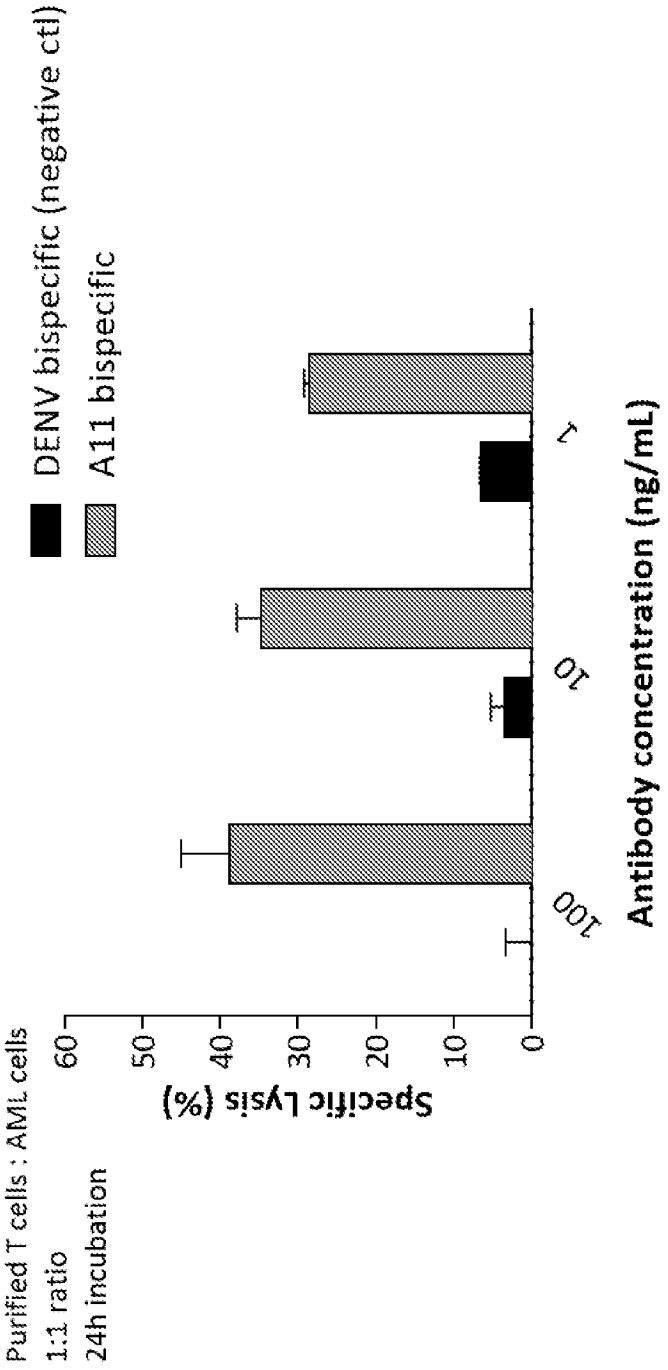
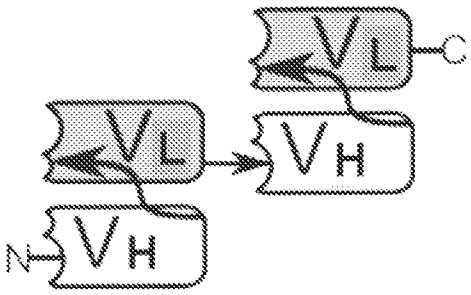

DENV bispecific (negative ctl)
A11 bispecific
Purified T cells : AML cells
1:1 ratio
24h incubation
Specific Lysis (%)
0
10
20
30
40
50
60
100
10
1
Antibody concentration (ng/mL)

Figure 16



tandem di-scFv

Figure 17

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SG2015/050415

A. CLASSIFICATION OF SUBJECT MATTER

See Supplemental Box

According to International Patent Classification (IPC)

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

A61K C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

CAplus/BIOSIS/EMBASE/MEDLINE/FamPat/EPODOC/WPIAP: TIM-3, T cell immunoglobulin mucin 3, HAVCR2, antibodies, immunoglobulin, T cell dysfunction, CD3 and related terms.
STN DGENE: SEQ Nos. 45 and 50.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 2012/0189617 A1 (TAKAYANAGI S-I. ET AL) 26 July 2012 Paragraphs [0022], [0057], [0395]-[0400]; Examples 8, 9, 17 and 18.	1-92
X	WO 2013/006490 A2 (CELLERANT THERAPEUTICS, INC.) 10 January 2013 Paragraph [0007]; Examples 1, 7, 8 and 10.	1-92
X	CN 103936853 A (INSTITUTE OF BASIC MEDICAL SCIENCES, THE ACADEMY OF MILITARY MEDICAL SCIENCES) 23 July 2014 Paragraphs [0003], [0005], [0006]; Examples 1, 4, 7 and 8; Table 1.	1-92
X	CN 102492038 A (INSTITUTE OF BASIC MEDICAL SCIENCES, THE ACADEMY OF MILITARY MEDICAL SCIENCES) 13 June 2012 Paragraphs [0007] and [0008]; Examples 1 and 2.	1-92
A	WO 2011/159877 A2 (THE BRIGHAM AND WOMEN'S HOSPITAL, INC.) 22 December 2011 Whole document, specifically paragraphs [0005], [0122], [0123] and Examples.	-

☒ Further documents are listed in the continuation of Box C. ☒ See patent family annex.

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"A" document defining the general state of the art which is not considered to be of particular relevance

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"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

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"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
11/12/2015 *(day/month/year)*	21/12/2015 *(day/month/year)*

Name and mailing address of the ISA/SG

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INTERNATIONAL SEARCH REPORT

International application No.
PCT/SG2015/050415

C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	LUM L.G. AND THAKUR A., Targeting T cells with Bispecific Antibodies for Cancer Therapy. *BioDrugs*, 1 December 2011, Vol. 25, No. 6, pages 365-379 (NIH Public Access Author Manuscript in PMC) [Retrieved on 2015-12-14 from http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3792709] Whole document, particularly Sections 3, 4.2 and 6.4; Table III	-
A	WO 2014/116846 A2 (ABBVIE, INC) 31 July 2014 Whole document specifically, page 2, paragraph 1, page 3, paragraphs 1-4, page 18, paragraph 3.	-
P,X	WO 2015/117002 A1 (NOVARTIS AG ET AL) 6 August 2015 Whole document.	1-92
P,X	CN 104592388 A (CHINESE PLA GENERAL HOSPITAL) 6 May 2015 Whole document, specifically, page 2, paragraphs 2-3, page 18, paragraph 1; Examples 1 and 8-10.	1-92
P,X	WO 2015/103072 A1 (EPIMAB BIOTHERAPEUTICS) 9 July 2015 Whole document, specifically paragraphs [0016], [0029].	1-92

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SG2015/050415

Box No. I Nucleotide and/or amino acid sequence(s) (Continuation of item 1.c of the first sheet)

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application, the international search was carried out on the basis of a sequence listing:

   a. ☒ forming part of the international application as filed:

      ☒ in the form of an Annex C/ST.25 text file.

      ☐ on paper or in the form of an image file.

   b. ☐ furnished together with the international application under PCT Rule 13*ter*.1(a) for the purposes of international search only in the form of an Annex C/ST.25 text file.

   c. ☐ furnished subsequent to the international filing date for the purposes of international search only:

      ☐ in the form of an Annex C/ST.25 text file (Rule 13*ter*.1(a)).

      ☐ on paper or in the form of an image file (Rule 13*ter*.1(b) and Administrative Instructions, Section 713).

2. ☐ In addition, in the case that more than one version or copy of a sequence listing has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

   Since only one version or copy of a sequence listing has been filed or furnished, the statements under item 2 are not required.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SG2015/050415

Supplemental Box
(Classification of Subject Matter)

Int. Cl.
A61K 39/395 (2006.01)
C07K 16/28 (2006.01)
A61P 31/00 (2006.01)
A61P 35/00 (2006.01)
A61P 37/00 (2006.01)

INTERNATIONAL SEARCH REPORT

Information on patent family members

International application No.
PCT/SG2015/050415

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 2012/189617 (A1)	26/7/2012	AU2011262758 (A1) CA2814155 (A1) CN103079644 (A) EP2581113 (A1) KR20130132695 (A) TW201207397 (A) US2014044728 (A1) WO2011155607 (A1)	10/1/2013 15/12/2011 1/5/2013 17/4/2013 5/12/2013 16/2/2012 13/2/2014 15/12/2011
WO 2013/006490 (A2)	10/1/2013	US2013022623 (A1) US2015086574 (A1)	24/1/2013 26/3/2015
CN 103936853 (A)	23/07/2014	NONE	
CN 102492038 (A)	13/06/2012	NONE	
WO 2011/159877 (A2)	22/12/2011	CA2802344 (A1) JP2013532153 (A) US2013156774 (A1)	22/12/2011 15/8/2013 20/6/2013
WO 2014/116846 (A2)	31/7/2014	EP2948475 (A2) US2014242077 (A1)	2/12/2015 28/8/2014
WO 2015/117002 (A1)	6/8/2015	US2015218274 (A1) UY35973 (A)	6/8/2015 31/8/2015
CN 104592388 (A)	06/05/2015	NONE	
WO 2015/103072 (A1)	09/07/2015	NONE	