on data described for the triomAb bispecific antibody in the prior art it was highly doubted that a CD3xTAA bispecific IgG of a fully human IgG1 format would be able to induce lytic anti-tumor activity in resting peripheral blood lymphocytes without the need for pre-activation of T cells. In addition, the available data for the BiTE® format suggested that a full length IgG molecule would be too large to create effective cytolytic synapses between tumor cells and effector cells. Surprisingly, we demonstrated that a fully human CD3xCLEC12A bispecific full length IgG1 was able to induce very efficient T cell mediated lysis of CLEC12A-positive HL60 AML tumor cells in vitro. In fact, effective lysis was mediated by resting T lymphocytes purified from PBMC without the need of prior activation of the T cells. Furthermore, we demonstrated that this lytic activity is not necessarily dependent on interactions with FcγR present on HL60 cells as this lytic activity was not affected by the presence of excess human IgG when the assay was performed in human serum containing media. This is the first time that a full length human IgG1 bispecific T cell engager antibody exerts efficient tumor cell lysis without the need of pre-activation of T cells or the need of active FcγR interactions. Effective lysis is achieved despite the relatively large size of the IgG1 when compared to BiTE® molecules. Remarkably, when CH2/lower hinge mutations were introduced in the CD3xCLEC12A bispecific IgG1 molecule to further decrease Fc receptor interactions, this still resulted in efficient tumor cell lysis by immune effector cells. A bispecific human IgG1 T cell engager antibody has advantages over current IgG that make use of the hybrid subclass combination mouse IgG2a/rat IgG2b, since a human IgG1 will be less immunogenic and can thus be applied for repeated therapy. In addition, a full length bispecific human IgG1 T cell engager antibody has advantages over immunoglobulin/like molecules such as DART™, TandAb® or BiTE® as the full length human IgG1 is not rapidly cleared from the circulation and dosing of patients will thus not require continuous infusion, which is more beneficial to patients.

[0026a] In one aspect, the present disclosure refers to a bispecific ~~immunoglobulin~~IgG antibody, wherein said bispecific IgG antibody~~immunoglobulin~~ comprises one arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ and a second arm that specifically recognizes an antigen on immune effector cells capable of recruiting such cells to an aberrant cell expressing CLEC12A ~~or CLL-1 or DCAL-2 or MICL or KLRL1~~, wherein

the arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ comprises:

- a heavy chain CDR1 sequence comprising the sequence SGYTFTSY and a heavy chain CDR2 sequence comprising the sequence IINPSGGS and a heavy chain CDR3 sequence comprising the sequence GTTGDWFDY and a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT; or
- a heavy chain CDR1 sequence comprising the sequence SGYTFTSY and a heavy chain CDR2 sequence comprising the sequence IINPSGGS and a heavy chain CDR3 sequence comprising the sequence GNYGDEFDY and a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT; or
- a heavy chain CDR1 sequence comprising the sequence SGYTFTGY and a heavy chain CDR2 sequence comprising the sequence WINPNSGG and a heavy chain CDR3 sequence comprising the sequence DGYFADAFDY and a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT~~; or~~
- ~~CDR sequences that deviate in no more than three amino acids from the recited CDR sequences~~.

[0026b] In another aspect, the present disclosure refers to a method for producing a bispecific IgG antibody~~immunoglobulin~~ as disclosed herein from a single cell, wherein said bispecific IgG antibody~~immunoglobulin~~ comprises two CH3 domains that are capable of forming an interface, said method comprising providing:

- a cell comprising a) a first nucleic acid sequence encoding an IgG antibody~~immunoglobulin~~ heavy chain that specifically recognizes CLEC12A and that comprises a 1st CH3 domain, and b) a second nucleic acid sequence encoding an IgG antibody ~~immunoglobulin~~ heavy chain that specifically recognizes an antigen on immune effector cells, preferably CD3, and that comprises a 2nd CH3 domain, wherein said nucleic acid sequences are provided with amino acid substitutions in the 1st and 2nd CH3 domains for preferential pairing of said 1st and 2nd CH3 domains, said method further comprising the step of culturing said cell and allowing for expression of said two nucleic acid sequences and harvesting said bispecific IgG antibody~~immunoglobulin~~ from the culture.

[0026c] In another aspect, the present disclosure refers to a pharmaceutical composition comprising a bispecific IgG antibody~~immunoglobulin~~ as disclosed herein and a pharmaceutically acceptable carrier.

[0026d] In another aspect, the present disclosure refers to a bispecific IgG antibody~~immunoglobulin~~ as disclosed herein for use as a pharmaceutical in the treatment of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).

[0026e] In another aspect, the present disclosure refers to use of a bispecific IgG antibody~~immunoglobulin~~ as disclosed herein in the preparation of a medicament for the treatment of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).

EMBODIMENTS

[0027] The invention provides a bispecific IgG antibody, wherein said bispecific IgG antibody comprises one arm that specifically recognizes CLEC12A or a functional equivalent thereof, and a second arm that specifically recognizes an antigen on immune effector cells capable of recruiting such cells to an aberrant cell expressing CLEC12A or said functional equivalent.

CLAIMS

1. A bispecific ~~immunoglobulin~~IgG antibody, wherein said bispecific ~~immunoglobulin~~ IgG antibody comprises one arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ and a second arm that specifically recognizes an antigen on immune effector cells capable of recruiting such cells to an aberrant cell expressing CLEC12A ~~or CLL-1 or DCAL-2 or MICL or KLRL1~~,

wherein the arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ comprises:

- a heavy chain CDR1 sequence comprising the sequence SGYTFTSY and a heavy chain CDR2 sequence comprising the sequence IINPSGGS and a heavy chain CDR3 sequence comprising the sequence GTTGDWFDY and a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT; or

- a heavy chain CDR1 sequence comprising the sequence SGYTFTSY and a heavy chain CDR2 sequence comprising the sequence IINPSGGS and a heavy chain CDR3 sequence comprising the sequence GNYGDEFDY and a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT; or

- a heavy chain CDR1 sequence comprising the sequence SGYTFTGY and a heavy chain CDR2 sequence comprising the sequence WINPNSGG and a heavy chain CDR3 sequence comprising the sequence DGYFADAFDY and a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT~~; or~~

~~- CDR sequences that deviate in no more than three amino acids from the recited CDR sequences~~.

~~2. The bispecific immunoglobulin according to claim 1, wherein said CDR sequences deviate in no more than two amino acids from the recited CDR sequences.~~

~~3. The bispecific immunoglobulin according to claim 1 or 2, wherein said CDR sequences deviate in no more than one amino acid from the recited CDR sequences.~~

24. The bispecific IgG antibody ~~immunoglobulin~~ according to ~~any one of~~ claim~~s~~ 1~~-3~~, wherein said immune effector cells comprise T cells.

35. The bispecific IgG antibody ~~immunoglobulin~~ according to claim 1 or 2~~any one of claims 1-4~~, wherein said antigen on said immune effector cells is CD3.

46. The bispecific IgG antibody ~~immunoglobulin~~ according to claim 35, wherein said IgG antibody ~~immunoglobulin~~ specifically recognizes CD3ε.

57. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-46, comprising a ~~wherein said~~ common light chain, preferably ~~is~~ a germline light chain, preferably the rearranged germline human kappa light chain IgVκ1-39*01/IGJκ1*01.

68. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-57, wherein said bispecific IgG antibody ~~immunoglobulin~~ is a human IgG antibody, preferably a human IgG1.

79. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-68, wherein the arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ comprises a variable heavy chain sequence ~~consisting of a sequence that is at least 90% identical to~~
QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGS TSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCAKGTTGDWFDYWGQGTLV TVSS.

810. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-68, wherein the arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ comprises a variable heavy chain sequence ~~consisting of a sequence that is at least 90%~~

~~identical to~~

EVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGS TSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARGNYGDEFDYWGQGTLV TVSS.

9~~11~~. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-6~~8~~, wherein the arm that specifically recognizes CLEC12A~~, or CLL-1 or DCAL-2 or MICL or KLRL1,~~ comprises a variable heavy chain sequence ~~consisting of a sequence that is at least 90% identical to~~

QVQLVQSGAEVKKPGASVKVSCKASGYTFTGYYMHWVRQAPGQGLEWMGWINPNSG GTNYAQKFQGRVTMTRDTSISTAYMELSRLRSDDTAVYYCARDGYFADAFDYWGQGT LVTVSS.

10~~12~~. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 3-9~~5-11~~, wherein the second arm that specifically recognizes CD3 comprises a heavy chain CDR1 sequence consisting of the sequence SYGMH and a heavy chain CDR2 sequence consisting of the sequence IIWYSGSKKNYADSVKG and a heavy chain CDR3 sequence consisting of the sequence GTGYNWFDP.

11~~13~~. The bispecific IgG antibody ~~immunoglobulin~~ according to claim 10~~12~~, wherein the second arm that specifically recognizes CD3 comprises a variable heavy chain sequence consisting of the sequence

QVQLVESGGGVVQPGRSLRLSCAASGFTFRSYGMHWVRQAPGKGLEWVAIIWYSGSK KNYADSVKGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCARGTGYNWFDPWGQGTLV TVSS.

~~14. The bispecific immunoglobulin according to any one of claims 1-14, wherein the first and the second arms further comprise a light chain CDR1 sequence comprising the sequence RASQSISSYLN and a light chain CDR2 sequence comprising the sequence AASSLQS and a light chain CDR3 sequence comprising the sequence QQSYSTPPT.~~

12~~15~~. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-11~~claim 14~~, wherein the first and the second arms comprise a variable light chain sequence ~~consisting of a sequence that is at least 90% identical to~~ DIQMTQSPSSLSASVGDRVTITCRASQSISSYLNWYQQKPGKAPKLLIYAASSLQSGVPSR FSGSGSGTDFTLTISSLQPEDFATYYCQQSYSTPPTFGQGTKVEIK.

13~~16~~. The bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-12~~15~~, wherein said bispecific IgG antibody ~~immunoglobulin~~ comprises mutated CH2 and/or lower hinge domains such that interaction of said bispecific IgG antibody ~~immunoglobulin~~ with Fcγ receptors is significantly reduced.

14~~17~~. The bispecific IgG antibody ~~immunoglobulin~~ according to claim 13~~16~~, wherein said mutated CH2 and/or lower hinge domains comprise at least one substitution at amino acids position 235 and/or 236.

15~~18~~. The bispecific IgG antibody ~~immunoglobulin~~ according to claim 13 or 14~~16 or 17~~, wherein said mutated CH2 and/or lower hinge domains comprise substitution L235G and/or G236R, preferably L235G and G236R.

16~~19~~. A method for producing a bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-15~~18~~ from a single cell, wherein said bispecific IgG antibody ~~immunoglobulin~~ comprises two CH3 domains that are capable of forming an interface, said method comprising providing:

- a cell comprising a) a first nucleic acid sequence encoding an IgG antibody ~~immunoglobulin~~ heavy chain that specifically recognizes CLEC12A and that comprises a 1st CH3 domain, and b) a second nucleic acid sequence encoding an IgG antibody ~~immunoglobulin~~ heavy chain that specifically recognizes an antigen on immune effector cells, preferably CD3, and that comprises a 2nd CH3 domain, wherein said nucleic acid sequences are provided with amino acid substitutions in the 1st and 2nd CH3 domains for preferential pairing of said 1st and 2nd CH3 domains, said method further comprising the step of culturing said cell and allowing for

expression of said two nucleic acid sequences and harvesting said bispecific IgG antibody ~~immunoglobulin~~ from the culture.

17~~20~~. A method according to claim 16~~19~~, wherein said cell has a third nucleic acid sequence encoding a common light chain, preferably the rearranged germline human kappa light chain IgVκ1-39*01/IGJκ1*01.

18~~21~~. A method according to claim 16 or 17~~19 or 20~~ for producing a bispecific IgG antibody ~~immunoglobulin~~, wherein said first CH3 domain comprises the amino acid substitutions L351K and T366K and wherein said second CH3 domain comprises the amino acid substitutions L351D and L368E, said method further comprising the step of culturing said cell and allowing for expression of said nucleic acid sequences and harvesting said bispecific IgG antibody ~~immunoglobulin~~ from the culture.

19~~22~~. A pharmaceutical composition comprising a bispecific IgG antibody ~~immunoglobulin~~ of any one of claims 1-15~~18~~ and a pharmaceutically acceptable carrier.

20~~23~~. A bispecific IgG antibody ~~immunoglobulin~~ of any one of claims 1-15~~18~~ for use as a pharmaceutical in the treatment of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).

21~~24~~. Use of a bispecific IgG antibody ~~immunoglobulin~~ according to any one of claims 1-15~~18~~ in the preparation of a medicament for the treatment of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).