et al. Molecular Immunology 2006 (43) 763–771) which is also believed to be linked to the small size of the BiTE® format. If size matters, this would suggest that larger molecules such as intact IgG would be too large to form effective cytolytic synapses. The CD3xCD19 BiTE®, blinatumomab, has demonstrated remarkable clinical efficacy in refractory non-Hodgkin lymphoma and acute lymphatic leukemia patients (Bargou et al. 2008 Science 321:974). Although the CD3xCD19 BiTE® displays very efficient tumor cell lysis at low levels in vitro, administration of this bispecific format to patients is associated with significant challenges. Due to their small size, BiTE®s are rapidly cleared from the circulation and dosing of patients thus requires continuous infusion. As the dosing regimen has an overall duration of more than 2 months, this treatment has a significant impact on the quality of life of the patients.

[0004] There thus remains a need for effective full length bispecific T cell engaging IgG molecules in eradicating aberrant cells that combine a long circulatory half-life upon intravenous administration without the need for continuous infusion without being immunogenic and with only limited side effects.

SUMMARY

[0004a] In one aspect, there is provided a bispecific human IgG full length antibody, wherein said bispecific IgG antibody comprises one arm that specifically recognizes CLEC12A, or CLL 1 or DCAL 2 or MICL or KLRL1, ~~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 CLEC12A or CLL 1 or DCAL 2 or MICL or KLRL1 ~~or said functional equivalent~~.

[0004b] In another aspect, there is provided a method for producing a bispecific human IgG full length antibody as defined herein from a single cell, wherein said bispecific IgG antibody comprises two CH3 domains that are capable of forming an interface, said method comprising providing:

- a cell having a) a first nucleic acid sequence encoding a IgG heavy chain that specifically recognizes CLEC12A and that contains a 1st CH3 domain, and b) a second nucleic acid sequence encoding a IgG heavy chain that specifically recognizes an antigen on immune effector cells, preferably CD3, and that contains a 2nd CH3 domain, wherein said nucleic acid sequences are provided with means for preferential pairing of said 1st and 2nd CH3 domains, said method

CLAIMS

1. A bispecific human IgG full length 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.

2. The bispecific IgG antibody according to claim 1, wherein said immune effector cells comprise T cells.

3. The bispecific IgG antibody according to claim 1 or 2, wherein said antigen on said immune effector cells is CD3.

4. The bispecific IgG antibody according to claim 3, wherein said antibody specifically recognizes CD3ε.

5. The bispecific IgG antibody according to any one of claims 1-4, wherein both arms comprise a common light chain.

6. The bispecific IgG antibody according to any one of claims 1-5, wherein said common light chain is a germline light chain, preferably the rearranged germline human kappa light chain IgVκ1-39*01/IGJκ1*01.

7. The bispecific IgG antibody according to any one of claims 1-6, wherein said bispecific antibody is a human IgG1.

8. The bispecific IgG antibody according to any one of claims 1-7, wherein the arm that specifically recognizes CLEC12A or a functional equivalent thereof comprises a heavy chain CDR1 sequence consisting of a sequence that is at least 90% identical to SGYTFTSY and a heavy chain CDR2 sequence consisting of a sequence that is at least 90% identical to IINPSGGS

and a heavy chain CDR3 sequence consisting of a sequence that is at least 90% identical to GTTGDWFDY.

9. The bispecific IgG antibody according to claim 8, wherein the arm that specifically recognizes CLEC12A or a functional equivalent thereof comprises a variable heavy chain sequence consisting of a sequence that is at least 90% identical to QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGS TSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCAKGTTGDWFDYWGQGTLV TVSS.

10. The bispecific IgG antibody according to any one of claims 1-7, wherein the arm that specifically recognizes CLEC12A or a functional equivalent thereof comprises a heavy chain CDR1 sequence consisting of a sequence that is at least 90% identical to SGYTFTSY and a heavy chain CDR2 sequence consisting of a sequence that is at least 90% identical to IINPSGGS and a heavy chain CDR3 sequence consisting of a sequence that is at least 90% identical to GNYGDEFDY.

11. The bispecific IgG antibody according to claim 10, wherein the arm that specifically recognizes CLEC12A or a functional equivalent thereof comprises a variable heavy chain sequence consisting of a sequence that is at least 90% identical to EVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGS TSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARGNYGDEFDYWGQGTLV TVSS.

12. The bispecific IgG antibody according to any one of claims 1-7, wherein the arm that specifically recognizes CLEC12A or a functional equivalent thereof comprises a heavy chain CDR1 sequence consisting of a sequence that is at least 90% identical to SGYTFTGY and a heavy chain CDR2 sequence consisting of a sequence that is at least 90% identical to WINPNSGG and a heavy chain CDR3 sequence consisting of a sequence that is at least 90% identical to DGYFADAFDY.

13. The bispecific IgG antibody according to claim 12, wherein the arm that specifically recognizes CLEC12A or a functional equivalent thereof comprises a variable heavy chain sequence consisting of a sequence that is at least 90% identical to QVQLVQSGAEVKKPGASVKVSCKASGYTFTGYYMHWVRQAPGQGLEWMGWINPNSG GTNYAQKFQGRVTMTRDTSISTAYMELSRLRSDDTAVYYCARDGYFADAFDYWGQGT LVTVSS.

14. The bispecific IgG antibody according to any one of claims 3-13, 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.

15. The bispecific IgG antibody according to claim 14, wherein the second arm that specifically recognizes CD3 comprises a variable heavy chain sequence consisting of the sequence QVQLVESGGGVVQPGRSLRLSCAASGFTFRSYGMHWVRQAPGKGLEWVAIIWYSGSK KNYADSVKGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCARGTGYNWFDPWGQGTLV TVSS.

16. The bispecific IgG antibody according to any one of claims 1-15, wherein the first and the second arms further comprise a light chain CDR1 sequence consisting of a sequence that is at least 90% identical to RASQSISSYLN and a light chain CDR2 sequence consisting of a sequence that is at least 90% identical to AASSLQS and a light chain CDR3 sequence consisting of a sequence that is at least 90% identical to QQSYSTPPT.

17. The bispecific IgG antibody according to claim 16, 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.

18. The bispecific IgG antibody according to any one of claims 1-17, wherein said bispecific IgG antibody has mutated CH2 and/or lower hinge domains such that interaction of said bispecific IgG antibody with Fcγ receptors is significantly reduced.

19. The bispecific IgG antibody according to claim 18, wherein said mutated CH2 and/or lower hinge domains comprise at least one substitution at amino acids position 235 and/or 236 (numbering according to Kabat).

20. The bispecific IgG antibody according to claim 18 or 19, wherein said mutated CH2 and/or lower hinge domains comprise substitution L235G and/or G236R, preferably L235G and G236R.

21. A method for producing a bispecific human IgG full length antibody according to any one of claims 1-20 from a single cell, wherein said bispecific IgG antibody comprises two CH3 domains that are capable of forming an interface, said method comprising providing:
- a cell having a) a first nucleic acid sequence encoding a IgG heavy chain that specifically recognizes CLEC12A and that contains a 1st CH3 domain, and b) a second nucleic acid sequence encoding a IgG heavy chain that specifically recognizes an antigen on immune effector cells, preferably CD3, and that contains 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 from the culture.

22. A method according to claim 21, 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.

23. A method according to claim 21 or 22 for producing a bispecific IgG1 antibody, wherein said first CH3 domain comprises the amino acid substitutions L351K and T366K (numbering according to Kabat) 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 antibody from the culture.

24. A pharmaceutical composition comprising a bispecific human IgG full length antibody of any one of claims 1-20 and a pharmaceutically acceptable carrier.

25. A bispecific human IgG full length antibody of any one of claims 1-20 for use as a pharmaceutical in the treatment of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).

26. Use of a bispecific human IgG ful length antibody according to any one of claims 1-20 in the preparation of a medicament for the treatment or prevention of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).

CLAIMS

1. A bispecific human IgG full length antibody, wherein said bispecific 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.

2. The bispecific IgG antibody according to claim 1, wherein said immune effector cells comprise T cells.

3. The bispecific IgG antibody according to claim 1 or 2, wherein said antigen on said immune effector cells is CD3.

4. The bispecific IgG antibody according to claim 3, wherein said antibody specifically recognizes CD3ε.

5. The bispecific IgG antibody according to any one of claims 1-4, wherein both arms comprise a common light chain.

6. The bispecific IgG antibody according to any one of claims 1-5, wherein said common light chain is a germline light chain, preferably the rearranged germline human kappa light chain IgVκ1-39*01/IGJκ1*01.

7. The bispecific IgG antibody according to any one of claims 1-6, wherein said bispecific antibody is a human IgG1.

8. The bispecific IgG antibody according to any one of claims 1-7, wherein the arm that specifically recognizes CLEC12A, or CLL 1 or DCAL 2 or MICL or KLRL1, comprises a heavy chain CDR1 sequence consisting of a sequence that is at least 90% identical to SGYTFTSY and a heavy chain CDR2 sequence consisting of a sequence that is at least 90% identical to IINPSGGS and a heavy chain CDR3 sequence consisting of a sequence that is at least 90% identical to GTTGDWFDY.

9. The bispecific IgG antibody according to claim 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 QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGS TSYAQKFQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCAKGTTGDWFDYWGQGTLV TVSS.

10. The bispecific IgG antibody according to any one of claims 1-7, wherein the arm that specifically recognizes CLEC12A, or CLL 1 or DCAL 2 or MICL or KLRL1, comprises a heavy chain CDR1 sequence consisting of a sequence that is at least 90% identical to SGYTFTSY and a heavy chain CDR2 sequence consisting of a sequence that is at least 90% identical to IINPSGGS and a heavy chain CDR3 sequence consisting of a sequence that is at least 90% identical to GNYGDEFDY.

11. The bispecific IgG antibody according to claim 10, 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 EVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPSGGSTSYAQK FQGRVTMTRDTSTSTVYMELSSLRSEDTAVYYCARGNYGDEFDYWGQGTLVTVSS.

12. The bispecific IgG antibody according to any one of claims 1-7, wherein the arm that specifically recognizes CLEC12A, or CLL 1 or DCAL 2 or MICL or KLRL1, comprises a heavy chain CDR1 sequence consisting of a sequence that is at least 90% identical to SGYTFTGY and a heavy chain CDR2 sequence consisting of a sequence that is at least 90% identical to WINPNSGG and a heavy chain CDR3 sequence consisting of a sequence that is at least 90% identical to DGYFADAFDY.

13. The bispecific IgG antibody according to claim 12, 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.

14. The bispecific IgG antibody according to any one of claims 3-13, 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.

15. The bispecific IgG antibody according to claim 14, wherein the second arm that specifically recognizes CD3 comprises a variable heavy chain sequence consisting of the sequence QVQLVESGGGVVQPGRSLRLSCAASGFTFRSYGMHWVRQAPGKGLEWVAIIWYSGSK KNYADSVKGRFTISRDNSKNTLYLQMNSLRAEDTAVYYCARGTGYNWFDPWGQGTLV TVSS.

16. The bispecific IgG antibody according to any one of claims 1-15, wherein the first and the second arms further comprise a light chain CDR1 sequence consisting of a sequence that is at least 90% identical to RASQSISSYLN and a light chain CDR2 sequence consisting of a sequence that is at least 90% identical to AASSLQS and a light chain CDR3 sequence consisting of a sequence that is at least 90% identical to QQSYSTPPT.

17. The bispecific IgG antibody according to claim 16, wherein the first and the second arms comprise a variable light chain sequence consisting of a sequence that is at least 90% identical to DIQMTQSPSSLSASVGDRVTITCRASQSISSYLNWYQQKPGKAPKLLIYAASSLQSGVPSRFSGSG SGTDFTLTISSLQPEDFATYYCQQSYSTPPTFGQGTKVEIK.

18. The bispecific IgG antibody according to any one of claims 1-17, wherein said bispecific IgG antibody has mutated CH2 and/or lower hinge domains such that interaction of said bispecific IgG antibody with Fcγ receptors is significantly reduced.

19. The bispecific IgG antibody according to claim 18, wherein said mutated CH2 and/or lower hinge domains comprise at least one substitution at amino acids position 235 and/or 236 (numbering according to Kabat).

20. The bispecific IgG antibody according to claim 18 or 19, wherein said mutated CH2 and/or lower hinge domains comprise substitution L235G and/or G236R, preferably L235G and G236R.

21. A method for producing a bispecific human IgG full length antibody according to any one of claims 1-20 from a single cell, wherein said bispecific IgG antibody comprises two CH3 domains that are capable of forming an interface, said method comprising providing:
- a cell having a) a first nucleic acid sequence encoding a IgG heavy chain that specifically recognizes CLEC12A and that contains a 1st CH3 domain, and b) a second nucleic acid sequence encoding a IgG heavy chain that specifically recognizes an antigen on immune effector cells, preferably CD3, and that contains 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 from the culture.

22. A method according to claim 21, 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.

23. A method according to claim 21 or 22 for producing a bispecific IgG1 antibody, wherein said first CH3 domain comprises the amino acid substitutions L351K and T366K (numbering according to Kabat) 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 antibody from the culture.

24. A pharmaceutical composition comprising a bispecific human IgG full length antibody of any one of claims 1-20 and a pharmaceutically acceptable carrier.

25. A bispecific human IgG full length antibody of any one of claims 1-20 for use as a pharmaceutical in the treatment of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).

26. Use of a bispecific human IgG ful length antibody according to any one of claims 1-20 in the preparation of a medicament for the treatment or prevention of myelodysplastic syndrome (MDS), chronic myelogenous leukemia (CML) or preferably acute myeloid leukemia (AML).