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ABSTRACT

Antibodies and antigen-binding fragments of antibodies that bind GCC are disclosed. The antibodies bind an extracellular domain of GCC and can be internalized. In some embodiments, the antibodies are humanized, chimeric or human. Nucleic acids and vectors encoding the antibodies or portions thereof, recombinant cells that contain the nucleic acids, and compositions comprising the antibodies or antigen-binding fragments are also disclosed. The invention also provides therapeutic and diagnostic methods utilizing the antibodies and antigen-binding fragments provided herein.

What is claimed is:

1. An anti-GCC antibody molecule, wherein the light chain variable region and the heavy chain variable region of the anti-GCC antibody molecule are selected from: the light and heavy chain sequences disclosed in Table 2 for any one of the following antibodies:

5F9;

Ab 229; or

3G1.

2. The anti-GCC antibody molecule, of claim 1, wherein said anti-GCC antibody molecule is an IgG1 antibody.

3. The anti-GCC antibody molecule of claim 1, wherein said anti-GCC antibodymolecule is not conjugated to a therapeutic agent.

4. The anti-GCC antibody molecule of claim 1, wherein said anti-GCC antibodymolecule is conjugated to a therapeutic agent.

5. The anti-GCC antibody molecule of claim 1, wherein said anti-GCC antibody molecule is conjugated to a detectable label.

6. An anti-GCC antibody molecule, comprising light chain CDR1, CDR2, and CDR3, and heavy chain CDR1, CDR2, and CDR3, disclosed in Table 5 for any one of the following antibodies:

5F9;

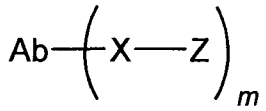
Ab 229; or

3G1.

7. The anti-GCC antibody molecule of claim 6, further comprising human or human derived light and heavy variable region frameworks.

8. The anti-GCC antibody molecule of claim 7, wherein said anti-GCC antibody is an IgG1 antibody.

9. An immunoconjugate of formula (*I*):



(*I*)

or a pharmaceutically acceptable salt thereof, wherein:

Ab is an anti-GCC antibody molecule of claim 1;

X is a linker moiety which connects Ab and Z;

Z is a therapeutic agent or a label; and

m is an integer from 1 to 15.

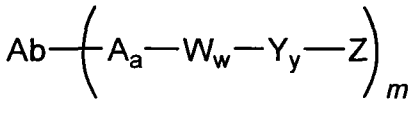
10. The immunoconjugate of claim 9, wherein Z is a detectable label.

11. The immunoconjugate of claim 9, wherein –Z is a maytansine or an auristatin.

12. The immunoconjugate of claim 9, wherein -Z is DM1 or DM4.

13. The immunoconjugate of claim 9, wherein -Z is MMAE or MMAF.

14. The immunoconjugate of claim 9, wherein the linker –X- has the formula $-A_a-W_w-Y_y-$, and the immunoconjugate is characterized by formula (*II*):



(*II*)

or a pharmaceutically acceptable salt thereof, wherein:

-A- is a Stretcher unit;

a is 0 or 1;

each –W- independently is an Amino Acid unit;

w is an integer ranging from 0 to 12;

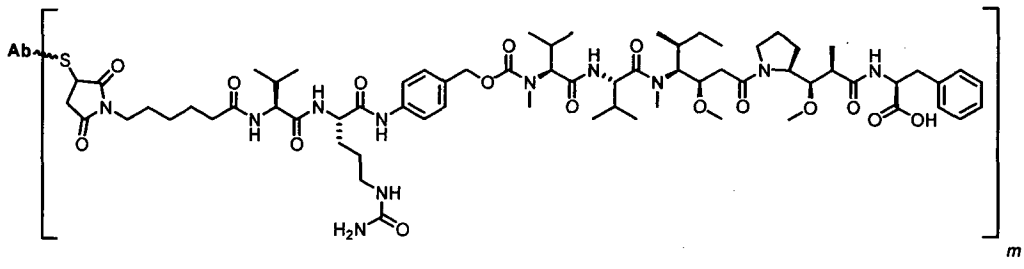
-Y- is a self-immolative Spacer unit;

y is 0, 1, or 2;

Z is a therapeutic agent or label; and

m ranges from about 1 to about 15.

15. An immunoconjugate of formula (*I-4*):



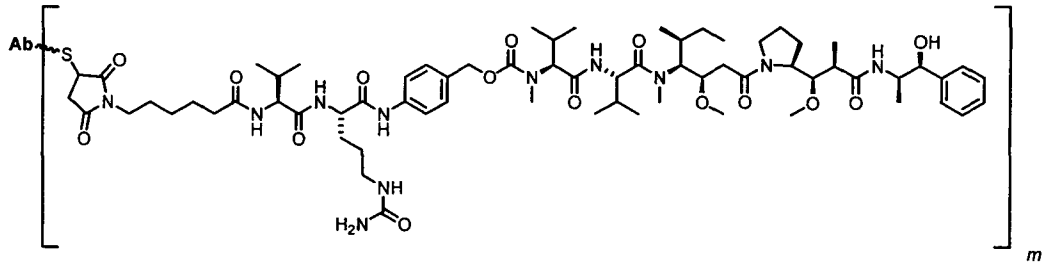
(*I-4*)

or a pharmaceutically acceptable salt thereof, wherein:

Ab is 5F9; and

m is an integer from 1 to 8.

16. An immunoconjugate of formula (*I-5*):



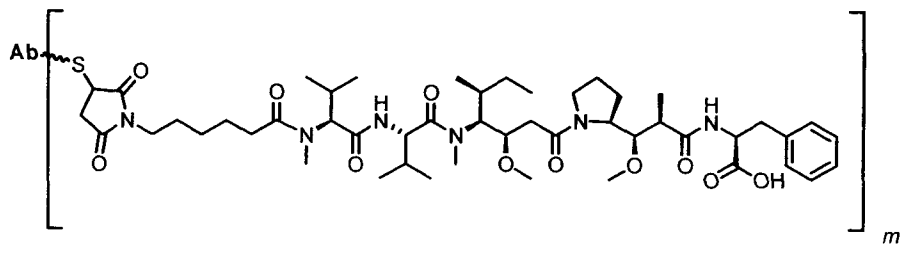
(*I-5*)

or a pharmaceutically acceptable salt thereof, wherein:

Ab is 5F9; and

m is an integer from 1 to 8.

17. An immunoconjugate of formula (*I-7*):



(*I-7*)

or a pharmaceutically acceptable salt thereof, wherein:

Ab is 5F9; and

m is an integer from 1 to 8.

18. The immunoconjugate of claim 15, wherein *m* is from 1 to 3.

19. The immunoconjugate of claim 15, wherein *m* is from 3 to 5.

20. The immunoconjugate of claim 16, wherein *m* is from 3 to 5.

21. The immunoconjugate of claim 18, wherein *m* is from 3 to 5.

22. A method of treating a subject for colon cancer, comprising administering to said subject, a therapeutically effective amount of an antibody molecule of claim 1 or 6, thereby treating said subject.

23. A method of treating a subject for colon cancer, comprising administering to said subject a therapeutically effective amount of an immunoconjugate of any one of claims 9-17.

24. Isolated nucleic acid sequences that encode an antibody molecule of claim 1 or 6.

25. A cell comprising the isolated nucleic acid sequences of claim 24.

26. A method of producing an antibodymolecule of claim 1 or 6, comprising culturing the cell of claim 25under conditions that allow production of an antibody molecule, thereby producing the antibody molecule of claim 1 or 6.

27. A vector comprising one or both of the light chain and heavy chain of an antibody molecule of claim 1 or 6.

28. A method of detecting a GCC molecule comprising contacting the molecule with an antibody molecule of claim 1 or 6 and determining if said antibody molecule, binds to said GCC molecule.

Dated this 26/03/2012

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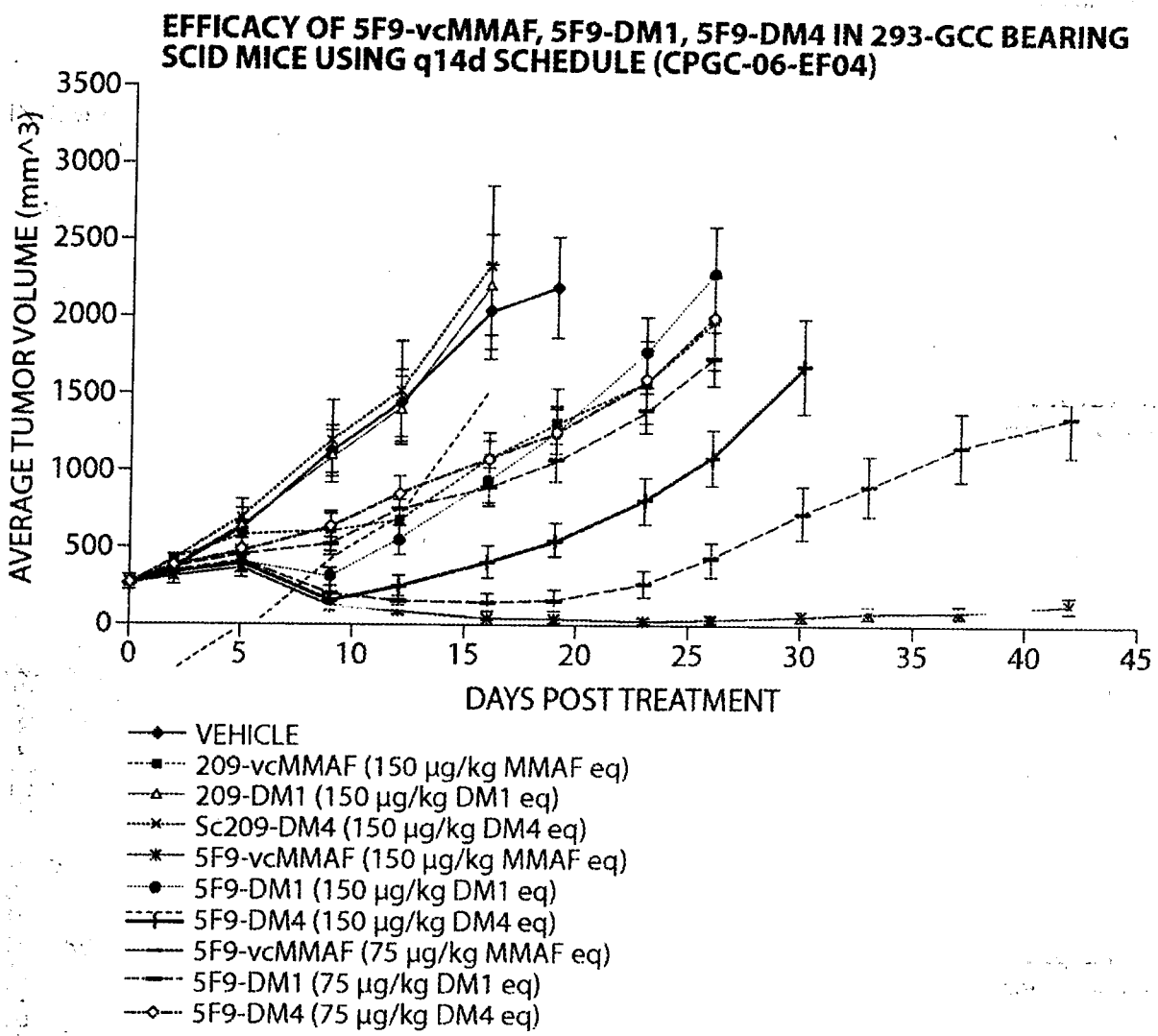

Fig. 1

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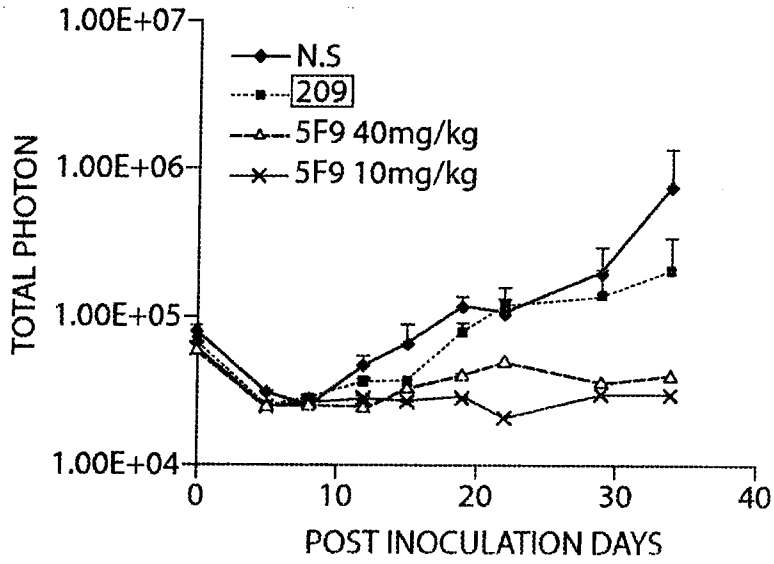
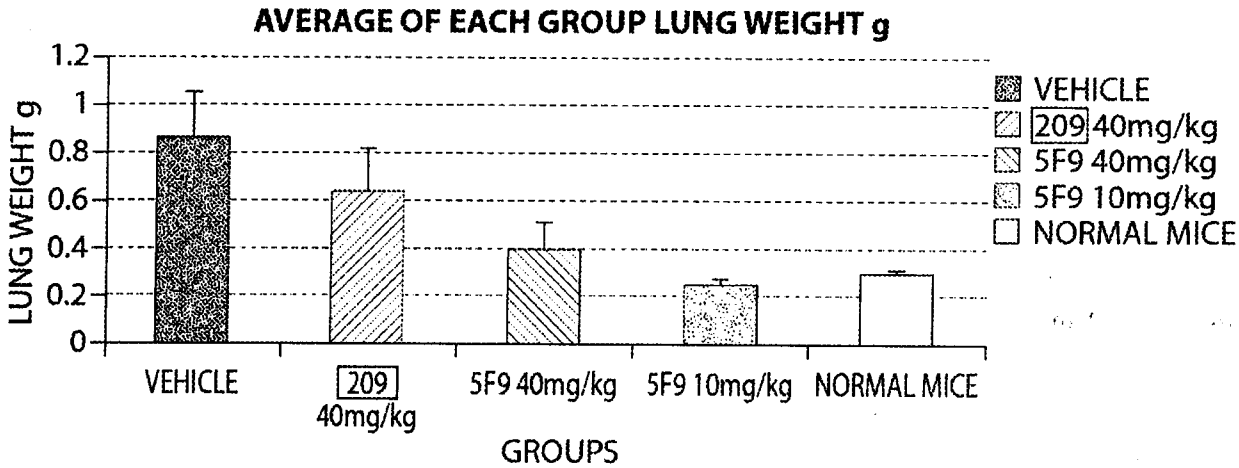


Fig. 2



T TEST: VEHICLE v.s 209 40mg/kg P=0.4
VEHICLE v.s 5F9 40mg/kg P=<0.05
VEHICLE v.s 5F9 10mg/kg P=<0.01

Fig. 3

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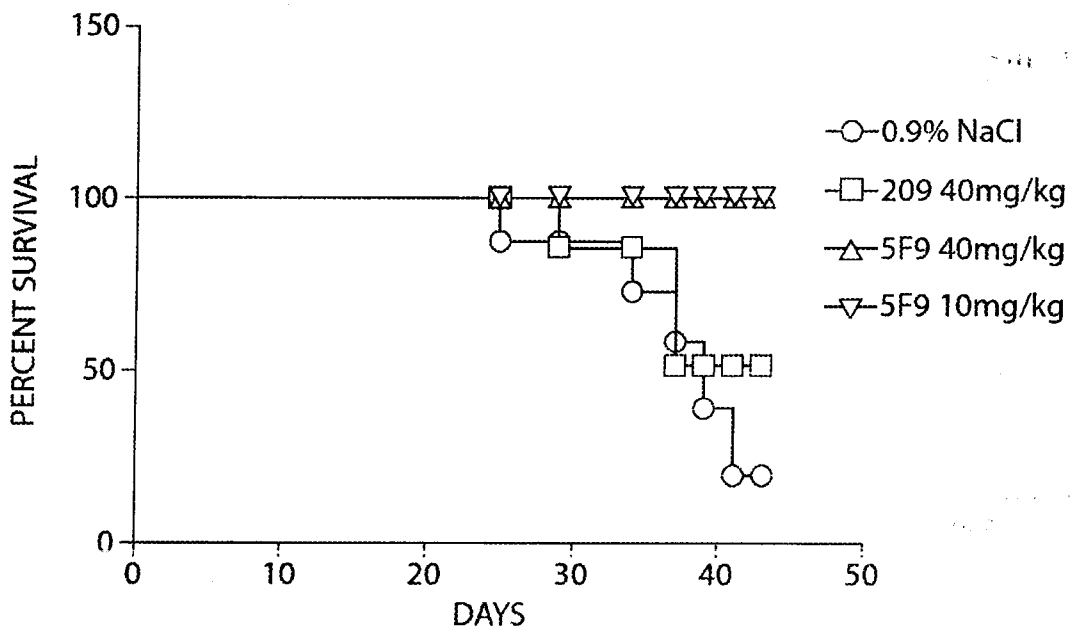

Fig. 4

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ANTI-GCC ANTIBODY MOLECULES AND RELATED COMPOSITIONS AND METHODS

RELATED APPLICATIONS

[001] The present application claims the benefit of U.S. Provisional Application Serial No. 61/254,474, filed October 23, 2009. The entire content of U.S. Provisional Application Serial No. 61/254,474 is incorporated herein by this reference.

FIELD OF INVENTION

[002] The invention relates to antibody molecules which bind GCC, as well as to related molecules, e.g., nucleic acids which encode such antibody molecules, compositions, and related methods, e.g., therapeutic and diagnostic methods.

BACKGROUND

[003] Guanylyl cyclase C (GCC) is a transmembrane cell surface receptor that functions in the maintenance of intestinal fluid, electrolyte homeostasis and cell proliferation, see, e.g., Carrithers et al., *Proc. Natl. Acad. Sci. USA* 100:3018-3020 (2003). GCC is expressed at the mucosal cells lining the small intestine, large intestine and rectum (Carrithers et al., *Dis Colon Rectum* 39: 171-181 (1996)). GCC expression is maintained upon neoplastic transformation of intestinal epithelial cells, with expression in all primary and metastatic colorectal tumors (Carrithers et al., *Dis Colon Rectum* 39: 171-181 (1996); Buc et al. *Eur J Cancer* 41: 1618-1627 (2005); Carrithers et al., *Gastroenterology* 107: 1653-1661(1994)).

SUMMARY

[004] The inventors have discovered numerous anti-GCC antibodies, including both human and murine antibodies. Accordingly, in one aspect, the invention features an anti-GCC antibody molecule, as disclosed herein. The anti-GCC antibody molecules are useful as naked antibody molecules and as components of immunoconjugates. Accordingly, in another aspect, the invention features immunoconjugates comprising an anti-GCC antibody molecule and a therapeutic agent or label. The invention also features pharmaceutical compositions comprising the anti-GCC antibody molecules and immunoconjugates described herein. The invention also features methods of using the anti-GCC antibody molecules and immunoconjugates described herein for detection of GCC and of cells or tissues that express GCC; for diagnosis, prognosis, imaging, or staging of a GCC-mediated disease; for modulating an activity or function of a GCC protein; and for treatment of a GCC-mediated disease, as described herein. In another aspect, the invention also features isolated and/or recombinant nucleic acids encoding anti-GCC antibody molecule amino acid sequences, as well as vectors and host cells comprising such nucleic acids, and methods for producing anti-GCC antibody molecules.

[005] All publications, patent applications, patents and other references mentioned herein are incorporated by references in their entirety.

[006] Other features, objects, and advantages of the invention(s) disclosed herein will be apparent from the description and drawings, and from the claims.

DESCRIPTION OF THE FIGURES

[007] Figure 1 depicts tumor growth in 293-GCC#2 bearing SCID mice treated with 5F9vc-MMAF, -DM1, and -DM4 on a q14d schedule.

[008] Figure 2 depicts lung weight of mice treated with 0.9%NaCl; 209 antibody at 40mg/kg; or 5F9 antibody at 10 or 40mg/kg on day 41 p.i.

[009] Figure 3 depicts the survival curve of CT26-hGCC tumor-bearing mice treated with 5F9 antibody.

[010] Figure 4 depicts ELISA binding assays to test antibody cross-reactivity of GCC orthologs.

DETAILED DESCRIPTION

Guanylyl Cyclase C

[011] Guanylyl cyclase C (GCC) (also known as STAR, ST Receptor, GUC2C, and GUCY2C) is a transmembrane cell surface receptor that functions in the maintenance of intestinal fluid, electrolyte homeostasis and cell proliferation (Carrithers et al., *Proc Natl Acad Sci U S A* 100: 3018-3020 (2003); Mann et al., *Biochem Biophys Res Commun* 239: 463-466 (1997); Pitari et al., *Proc Natl Acad Sci U S A* 100: 2695-2699 (2003)); GenBank Accession No. NM_004963, each of which is incorporated herein by reference). This function is mediated through binding of guanylin (Wiegand et al. FEBS Lett. 311:150-154 (1992)). GCC also is a receptor for heat-stable enterotoxin (ST, e.g., having an amino acid sequence of NTFYCCELCCNPACAGCY, SEQ ID NO:316) which is a peptide produced by *E. coli*, as well as other infectious organisms (Rao, M.C. *Ciba Found. Symp.* 112:74-93 (1985); Knoop F.C. and Owens, M. *J. Pharmacol. Toxicol. Methods* 28:67-72 (1992)). Binding of ST to GCC activates a signal cascade that results in enteric disease, e.g., diarrhea.

[012] Nucleotide sequence for human GCC (GenBank Accession No. NM_004963):

1 atgaagacgt tgctgttgga cttggctttg tggtcactgc tcttccagcc cgggtggctg

61 tcctttagtt cccaggtgag tcagaactgc cacaatggca gctatgaaat cagcgtcctg

121 atgatgggca actcagcctt tgcagagccc ctgaaaaact tggaagatgc ggtgaatgag

181 gggctggaaa tagtgagagg acgtctgcaa aatgctggcc taaatgtgac tgtgaacgct

241 actttcatgt attcggatgg tctgattcat aactcaggcg actgccggag tagcacctgt

301 gaaggcctcg acctactcag gaaaatttca aatgcacaac ggatgggctg tgtcctcata

361 gggccctcat gtacatactc caccttccag atgtaccttg acacagaatt gagctacccc

421 atgatctcag ctggaagttt tggattgtca tgtgactata aagaaacctt aaccaggctg

481 atgtctccag ctagaaagtt gatgtacttc ttggttaact tttggaaaac caacgatctg

541 cccttcaaaa cttattcctg gagcacttcg tatgtttaca agaatggtac agaaactgag

601 gactgtttct ggtaccttaa tgctctggag gctagcgttt cctatttctc ccacgaactc

661 ggctttaagg tggtgttaag acaagataag gagtttcagg atatcttaat ggaccacaac