CLAIMS

1. A pharmaceutical composition for the treatment of an epithelial tumor in a human, said pharmaceutical composition comprising a bispecific single chain antibody which has

(a) a first binding domain specifically binding to human CD3, and

(b) a second binding domain specifically binding to human CEA,

wherein said second binding domain comprises at least the amino acid sequence "$DX_1X_2X_3X_4FYFDY$" (SEQ ID NO. 65), wherein "X_1", "X_2", "X_3" or "X_4" represents any amino acid residue, and the amino acid residue "D" corresponds to Kabat position 95 of CDR-H3 of murine monoclonal antibody A5B7 and the amino acid residues "FYFDY" correspond to Kabat positions 100, 100a, 100b, 101, and 102, respectively, of CDR-H3 of murine monoclonal antibody A5B7.

2. The pharmaceutical composition of claim 1, wherein

- "X_1" represents "R" (Arginine), "F" (Phenylalanine), "M" (Methionine), "E" (Glutamic acid), or "T" (Threonine);
- "X_2" represents "G" (Glycine), "Y" (Tyrosine), "A" (Alanine), "D" (Aspartic acid), or "S" (Serine);
- "X_3" represents "L" (Leucine), "F" (Phenylalanine), "M" (Methionine), "E" (Glutamic acid), or "T" (Threonine); and
- "X_4" represents "R" (Arginine), "Y" (Tyrosine), "A" (Alanine), "D" (Aspartic acid), or "S" (Serine).

3. The pharmaceutical composition of claim 1 or 2, wherein said second binding domain specific for human CEA comprises at least the amino acid sequence "DRGLRFYFDY" (SEQ ID NO. 66) corresponding to Kabat positions 95 – 102 of the CDR-H3 of murine monoclonal antibody A5B7.

4. The pharmaceutical composition of any of claims 1 to 3, wherein said first binding domain specific for CD3 is located C-terminally.

5. The pharmaceutical composition of any of claims 1 to 4, wherein said binding domains are arranged in the order VH_{CEA}-VL_{CEA}-VH_{CD3}-VL_{CD3} or VL_{CEA}-VH_{CEA}-VH_{CD3}-VL_{CD3}.

6. The pharmaceutical composition of any of claims 1 to 5, wherein said second binding domain specific for human CEA comprises a CDR-H1 having the amino acid sequence "SYWMH" (SEQ ID NO. 68) and/or a CDR-H2 having the amino acid sequence "FIRNKANGGTTEYAASVKG" (SEQ ID NO. 67) or "FILNKANGGTTEYAASVKG" (SEQ ID NO. 145).

7. The pharmaceutical composition of any of claims 1 to 5, wherein said second binding domain specific for human CEA comprises a CDR-H1 having the amino acid sequence "TYAMH" (SEQ ID NO. 70) and/or a CDR-H2 having the amino acid sequence "LISNDGSNKYYADSVKG" (SEQ ID NO. 69).

8. The pharmaceutical composition of any of claims 1 to 7, wherein said second binding domain specific for human CEA comprises a CDR-L1 having the amino acid sequence "TLRRGINVGAYSIY" (SEQ ID NO. 73) and/or a CDR-L2 having the amino acid sequence "YKSDSDKQQGS" (SEQ ID NO. 72) and/or a CDR-L3 having the amino acid sequence "MIWHSGASAV" (SEQ ID NO. 71).

9. The pharmaceutical composition of claim 6, wherein the amino acid sequence of the VH region of the second binding domain specific for human CEA is SEQ ID NO. 60 or 146.

10. The pharmaceutical composition of claim 7, wherein the amino acid sequence of the VH region of the second binding domain specific for human CEA is SEQ ID NO. 58 or SEQ ID NO.62.

11. The pharmaceutical composition of claim 8, wherein the amino acid sequence of the VL region of the second binding domain specific for human CEA is SEQ ID NO. 64.

12. The pharmaceutical composition of any one of claims 1 to 11, wherein the V regions of the second binding domain specific for CEA are selected from the group consisting of:

(a) the VH region consists of the amino acid sequence shown in SEQ ID NO. 60 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64;

(b) the VH region consists of the amino acid sequence shown in SEQ ID NO. 146 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64

(c) the VH region consists of the amino acid sequence shown in SEQ ID NO. 58 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64;

(d) the VH region consists of the amino acid sequence shown in SEQ ID NO. 62 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64; and

(e) the VH region consists of the amino acid sequence shown in SEQ ID NO. 56 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64.

13. The pharmaceutical composition of any one of claims 1 to 12, wherein said bispecific single chain antibody comprises an amino acid sequence selected from the group consisting of:

(a) an amino acid sequence as depicted in any of SEQ ID NOs. 6, 8, 16, 18, 24, 26, 32, 34, 40, 42, 126, 130, 134 or 143;

(b) an amino acid sequence encoded by a nucleic acid sequence as shown in SEQ ID NOs. 5, 7, 15, 17, 23, 25, 31, 33, 39, 41, 125, 129, 133 or 142;

(c) an amino acid sequence encoded by a nucleic acid sequence hybridising under stringent conditions to the complementary nucleic acid sequence of (b);

(d) an amino acid sequence encoded by a nucleic acid sequence which is degenerate as a result of the genetic code to a nucleotide sequence of (b); and

(e) an amino acid sequence at least 85 % identical, more preferred at least 90 % identical, most preferred at least 95 % identical to the amino acid sequence of (a) or (b).

14. The pharmaceutical composition of any of claims 1 to 13, wherein said epithelial tumor is a gastrointestinal adenocarcinoma, a breast adenocarcinoma or a lung adenocarcinoma.

15. The pharmaceutical composition of claim 14, wherein said gastrointestinal adenocarcinoma is a colorectal, pancreatic, an oesophageal or a gastric adenocarcinoma.

16. The pharmaceutical composition of any of claims 1 to 15, wherein said pharmaceutical composition is for the treatment of progressive tumors, late stage tumors, tumor patients with high tumor load/burden, metastatic tumors, or tumor patients with a CEA serum concentration higher than 100 ng/ml.

17. The pharmaceutical composition of any of claims 1 to 16, wherein at least one of said first or second binding domains is chimeric, humanized, CDR-grafted, and/or deimmunized or human.

18. A pharmaceutical composition comprising a nucleic acid sequence encoding a bispecific single chain antibody as defined in any of claims 1 to 17.

19. A pharmaceutical composition comprising a vector which comprises a nucleic acid sequence as defined in claim 18.

20. The pharmaceutical composition of claim 19, wherein said vector further comprises a regulatory sequence which is operably linked to said nucleic acid sequence defined in claim 18.

21. The pharmaceutical composition of claim 19 or 20, wherein said vector is an expression vector.

22. A pharmaceutical composition comprising a host transformed or transfected with a nucleic acid defined in claim 18 or a vector defined in any of claims 19 to 21.

23. A pharmaceutical composition according to any of claims 1 to 22, further comprising a proteinaceous compound capable of providing an activation signal for immune effector cells.

24. A process for the production of a pharmaceutical composition according to any of claims 1 to 23, said process comprising culturing a host defined in claim 22 under conditions allowing the expression of the bispecific single chain antibody as defined in any of claims 1 to 17 and recovering the produced bispecific single chain antibody from the culture.

25. The pharmaceutical composition of any of claims 1 to 24, which is further comprising suitable formulations of carriers, stabilizers and/or excipients.

26. Use of a bispecific single chain antibody as defined in any of claims 1 to 17, a nucleic acid molecule as defined in claim 18, a vector as defined in any of claims 19 to 21, or a host as defined in claim 22, for the preparation of a pharmaceutical composition for the prevention, treatment or amelioration of an epithelial tumor in a human.

27. The use of claim 26, wherein said epithelial tumor is a gastrointestinal adenocarcinoma, a breast adenocarcinoma or a lung adenocarcinoma.

28. The use according to claim 27, wherein said gastrointestinal adenocarcinoma is a colorectal, pancreatic, an oesophageal or a gastric adenocarcinoma.

29. The use according to any of claims 1 to 28, wherein the pharmaceutical composition is for the treatment of progressive tumors, late stage tumors, tumor patients with high tumor load/burden, metastatic tumors, or tumor patients with a CEA serum concentration higher than 100 ng/ml.

30. The use according to any of claims 1 to 29, wherein said pharmaceutical composition is suitable to be administered in combination with an additional drug.

31. A method for the prevention, treatment or amelioration of an epithelial tumor in a subject in the need thereof, said method comprising the step of administration of an effective amount of a pharmaceutical composition of any of claims 1 to 17, or as produced according by the process of claim 24, over a sufficient period of time.

32. The method of claim 31, wherein said epithelial tumor is a gastrointestinal adenocarcinoma, a breast adenocarcinoma or a lung adenocarcinoma.

33. The method according to claim 32, wherein said gastrointestinal adenocarcinoma is a colorectal, pancreatic, an oesophageal or a gastric adenocarcinoma.

34. The method according to any of claims 31 to 33, wherein the pharmaceutical composition is for the treatment progressive tumors, late stage tumors, tumor patients with high tumor load/burden, metastatic tumors, or tumor patients with a CEA serum concentration higher than 100 ng/ml.

35. The method of any of claims 31 to 34, wherein said pharmaceutical composition is administered in combination with an additional drug.

36. The method of claim 35, wherein said drug is a non-proteinaceous compound or a proteinaceous compound.

37. The method of claim 36, comprising the administration of a proteinaceous compound capable of providing an activation signal for immune effector cells.

38. The method of claim 36 or 37, wherein said proteinaceous compound or non-proteinaceous compound is administered simultaneously or non-simultaneously with a bispecific single chain antibody as defined in any of

claims 1 to 17, a nucleic acid molecule as defined in claim 18, a vector as defined in any of claims 19 to 21, or a host as defined in claim 22.

39. The method of any one of claims 31 to 38, wherein said subject is a human.

40. A kit comprising a bispecific single chain antibody as defined in any of claims 1 to 17, a nucleic acid molecule as defined in claim 18, a vector as defined in any of claims 19 to 21, or a host as defined in claim 22.

41. The pharmaceutical composition of claim 16, the use of claim 29 or the method of claim 34, wherein said CEA serum concentration is determined by ELISA.

THE CLAIMS DEFINING THE INVENTION ARE AS FOLLOWS:

1. A bispecific single chain antibody for the treatment of an epithelial tumor in a human, said antibody comprising:

(a) a first binding domain specifically binding to human CD3; and

(b) a second binding domain specifically binding to human CEA,

wherein said second binding domain comprises at least the amino acid sequence "$DX_1X_2X_3X_4FYFDY$" (SEQ ID NO. 65), wherein "X_1", "X_2", "X_3" or "X_4" represents any amino acid residue, and the amino acid residue "D" corresponds to Kabat position 95 of CDR-H3 of murine monoclonal antibody A5B7 and the amino acid residues "FYFDY" correspond to Kabat positions 100, 100a, 100b, 101, and 102, respectively, of CDR-H3 of murine monoclonal antibody A5B7.

2. The bispecific single chain antibody of claim 1, wherein:

- "X_1" represents "R" (Arginine), "F" (Phenylalanine), "M" (Methionine), "E" (Glutamic acid), or "T" (Threonine);
- "X_2" represents "G" (Glycine), "Y" (Tyrosine), "A" (Alanine), "D" (Aspartic acid), or "S" (Serine);
- "X_3" represents "L" (Leucine), "F" (Phenylalanine), "M" (Methionine), "E" (Glutamic acid), or "T" (Threonine); and
- "X_4" represents "R" (Arginine), "Y" (Tyrosine), "A" (Alanine), "D" (Aspartic acid), or "S" (Serine).

3. The bispecific single chain antibody of claim 1 or claim 2, wherein said second binding domain specific for human CEA comprises at least the amino acid sequence "DRGLRFYFDY" (SEQ ID NO. 66) corresponding to Kabat positions 95 – 102 of the CDR-H3 of murine monoclonal antibody A5B7.

4. The bispecific single chain antibody of any one of claims 1 to 3, wherein said first binding domain specific for CD3 is located C-terminally.

5. The bispecific single chain antibody of any one of claims 1 to 4, wherein said binding domains are arranged in the order VH_{CEA}-VL_{CEA}-VH_{CD3}-VL_{CD3} or VL_{CEA}-VH_{CEA}-VH_{CD3}-VL_{CD3}.

6. The bispecific single chain antibody of any one of claims 1 to 5, wherein said second binding domain specific for human CEA comprises a CDR-H1 having the amino acid sequence "SYWMH" (SEQ ID NO. 68) and/or a CDR-H2 having the amino acid sequence "FIRNKANGGTTEYAASVKG" (SEQ ID NO. 67) or "FILNKANGGTTEYAASVKG" (SEQ ID NO. 145).

7. The bispecific single chain antibody of any one of claims 1 to 5, wherein said second binding domain specific for human CEA comprises a CDR-H1 having the amino acid sequence "TYAMH" (SEQ ID NO. 70) and/or a CDR-H2 having the amino acid sequence "LISNDGSNKYYADSVKG" (SEQ ID NO. 69).

8. The bispecific single chain antibody of any one of claims 1 to 7, wherein said second binding domain specific for human CEA comprises a CDR-L1 having the amino acid sequence "TLRRGINVGAYSIY" (SEQ ID NO. 73) and/or a CDR-L2 having the amino acid sequence "YKSDSDKQQGS" (SEQ ID NO. 72) and/or a CDR-L3 having the amino acid sequence "MIWHSGASAV" (SEQ ID NO. 71).

9. The bispecific single chain antibody of claim 6, wherein the amino acid sequence of the VH region of the second binding domain specific for human CEA is SEQ ID NO. 60 or SEQ ID NO. 146.

10. The bispecific single chain antibody of claim 7, wherein the amino acid sequence of the VH region of the second binding domain specific for human CEA is SEQ ID NO. 58 or SEQ ID NO. 62.

11. The bispecific single chain antibody of claim 8, wherein the amino acid sequence of the VL region of the second binding domain specific for human CEA is SEQ ID NO. 64.

12. The bispecific single chain antibody of any one of claims 1 to 11, wherein the V regions of the second binding domain specific for CEA are selected from the group consisting of:

(a) the VH region consists of the amino acid sequence shown in SEQ ID NO. 60 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64;

(b) the VH region consists of the amino acid sequence shown in SEQ ID NO. 146 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64;

(c) the VH region consists of the amino acid sequence shown in SEQ ID NO. 58 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64;

(d) the VH region consists of the amino acid sequence shown in SEQ ID NO. 62 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64; and

(e) the VH region consists of the amino acid sequence shown in SEQ ID NO. 56 and the VL region consists of the amino acid sequence shown in SEQ ID NO. 64.

13. The bispecific single chain antibody of any one of claims 1 to 12, comprising an amino acid sequence selected from the group consisting of:

(a) an amino acid sequence as depicted in any one of SEQ ID NOs. 6, 8, 16, 18, 24, 26, 32, 34, 40, 42, 126, 130, 134 or 143;

(b) an amino acid sequence encoded by a nucleic acid sequence as shown in any one of SEQ ID NOs. 5, 7, 15, 17, 23, 25, 31, 33, 39, 41, 125, 129, 133 or 142;

(c) an amino acid sequence encoded by a nucleic acid sequence hybridising under stringent conditions to a nucleic acid sequence

complementary to a nucleic acid sequence as shown in any one of SEQ ID NOs. 5, 7, 15, 17, 23, 25, 31, 33, 39, 41, 125, 129, 133 or 142;

(d) an amino acid sequence encoded by a nucleic acid sequence which is degenerate as a result of the genetic code to a nucleic acid sequence as shown in any one of SEQ ID NOs. 5, 7, 15, 17, 23, 25, 31, 33, 39, 41, 125, 129, 133 or 142; and

(e) an amino acid sequence at least 85 % identical, at least 90 % identical, or at least 95 % identical to the amino acid sequence of (a) or (b).

14. The bispecific single chain antibody of any one of claims 1 to 13, wherein said epithelial tumor is a gastrointestinal adenocarcinoma, a breast adenocarcinoma or a lung adenocarcinoma.

15. The bispecific single chain antibody of claim 14, wherein said gastrointestinal adenocarcinoma is a colorectal, pancreatic, an oesophageal or a gastric adenocarcinoma.

16. The bispecific single chain antibody of any one of claims 1 to 15, wherein said antibody is for the treatment of progressive tumors, late stage tumors, tumor patients with high tumor load/burden, metastatic tumors, or tumor patients with a CEA serum concentration higher than 100 ng/ml.

17. The bispecific single chain antibody of any one of claims 1 to 16, wherein at least one of said first or second binding domains is chimeric, humanized, CDR-grafted, and/or deimmunized or human.

18. A nucleic acid molecule encoding a bispecific single chain antibody as defined in any one of claims 1 to 17.

19. A vector comprising a nucleic acid molecule as defined in claim 18.

20. The vector of claim 19 further comprising a regulatory sequence which is operably linked to said nucleic acid molecule.

21. The vector of claim 19 or claim 20, wherein said vector is an expression vector.

22. A host transformed or transfected with a nucleic acid molecule defined in claim 18 or a vector defined in any one of claims 19 to 21.

23. A pharmaceutical composition comprising the bispecific single chain antibody of any one of claims 1 to 17, the nucleic acid molecule of claim 18, the vector of any one of claims 19 to 21, or the host of claim 22.

24. The pharmaceutical composition of claim 23, further comprising a proteinaceous compound capable of providing an activation signal for immune effector cells.

25. The pharmaceutical composition of claim 23 or claim 24, further comprising a carrier, a stabilizer and/or an excipient.

26. A process for the production of a bispecific single chain antibody, said process comprising culturing a host defined in claim 22 under conditions allowing the expression of the bispecific single chain antibody and recovering the produced bispecific single chain antibody from the culture.

27. Use of a bispecific single chain antibody as defined in any one of claims 1 to 17, a nucleic acid molecule as defined in claim 18, a vector as defined in any one of claims 19 to 21, or a host as defined in claim 22, in the manufacture of a medicament for the prevention, treatment or amelioration of an epithelial tumor in a human.

28. The use of claim 27, wherein said epithelial tumor is a gastrointestinal adenocarcinoma, a breast adenocarcinoma or a lung adenocarcinoma.

29. The use of claim 28, wherein said gastrointestinal adenocarcinoma is a colorectal, pancreatic, an oesophageal or a gastric adenocarcinoma.

30. The use of any one of claims 27 to 29, wherein the medicament is for the treatment of progressive tumors, late stage tumors, tumor patients with high tumor load/burden, metastatic tumors, or tumor patients with a CEA serum concentration higher than 100 ng/ml.

31. The use of any one of claims 27 to 30, wherein said medicament is suitable to be administered in combination with an additional drug.

32. A method for preventing, treating or ameliorating an epithelial tumor in a subject in need thereof, said method comprising the step of administering an effective amount of a bispecific single chain antibody of any one of claims 1 to 17, a bispecific single chain antibody produced by the process of claim 26, the nucleic acid molecule of claim 18, the vector of any one of claims 19 to 21, the host of claim 22, or the pharmaceutical composition of any one of claims 23 to 25 over a sufficient period of time.

33. The method of claim 32, wherein said epithelial tumor is a gastrointestinal adenocarcinoma, a breast adenocarcinoma or a lung adenocarcinoma.

34. The method of claim 33, wherein said gastrointestinal adenocarcinoma is a colorectal, pancreatic, an oesophageal or a gastric adenocarcinoma.

35. The method of any one of claims 32 to 34, wherein the antibody, the nucleic acid molecule, the vector, the host, or the pharmaceutical composition is for treating progressive tumors, late stage tumors, tumor patients with high tumor load/burden, metastatic tumors, or tumor patients with a CEA serum concentration higher than 100 ng/ml.

36. The method of any one of claims 32 to 35, wherein said antibody, the nucleic acid molecule, the vector, the host, or the pharmaceutical composition is administered in combination with an additional drug.

37. The method of claim 36, wherein said additional drug is a non-proteinaceous compound or a proteinaceous compound.

38. The method of claim 37, wherein said proteinaceous compound is capable of providing an activation signal for immune effector cells.

39. The method of claim 37 or claim 38, wherein said proteinaceous compound or non-proteinaceous compound is administered simultaneously or non-simultaneously with the bispecific single chain antibody, the pharmaceutical composition, the nucleic acid molecule, the vector, or the host.

40. The method of any one of claims 32 to 39, wherein said subject is a human.

41. A kit comprising a bispecific single chain antibody as defined in any one of claims 1 to 17, a nucleic acid molecule as defined in claim 18, a vector as defined in any one of claims 19 to 21, or a host as defined in claim 22.

42. The pharmaceutical composition of claim 16, the use of claim 30 or the method of claim 35, wherein said CEA serum concentration is determined by ELISA.

43. The bispecific single chain antibody of claim 1, the nucleic acid molecule of claim 18, the vector of claim 19, the host of claim 22, the pharmaceutical composition of claim 23, the process of claim 26, the use of claim 27, the method of claim 32, or the kit of claim 41, substantially as hereinbefore described with reference to the accompanying examples and/or figures.