

Novel therapeutic targets

Field of the invention

[0001] The invention relates to proteins and compositions for the treatment of cancers that express carbonic anhydrase IX (CAIX), and methods of use thereof.

Background of the invention

[0002] Molecular targeted radiotherapy (MTR) is an emerging approach for the treatment of cancers and includes the use of radiolabelled agents for binding to a molecular target.

[0003] However, the mere presence of a molecular target on a tumour is not always indicative that the cancer can be usefully treated using molecular targeted radiotherapy.

[0004] Accordingly, there is a need for new agents and methods for treating cancers for which there is currently an unmet need, or that have previously not been successfully targeted using MTR.

[0005] Reference to any prior art in the specification is not an acknowledgment or suggestion that this prior art forms part of the common general knowledge in any jurisdiction or that this prior art could reasonably be expected to be understood, regarded as relevant, and/or combined with other pieces of prior art by a skilled person in the art.

Summary of the invention

[0006] The present invention relates to reagents and methods for treating cancers that express the antigen carbonic anhydrase IX (CAIX).

[0007] The invention provides a method of treating, preventing or minimising progression of cancer in a subject, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer

- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby treating, preventing or minimising progression of the cancer in the subject.

[0008] In a further aspect, the present invention provides a method of minimising, reducing or preventing growth of a cancer tumour in a subject, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer

- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby minimising, reducing or preventing growth of the cancer tumour in the subject.

[0009] In a further aspect, the present invention provides a method of minimising, reducing or preventing metastasis of cancer in a subject, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer

- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby minimising, reducing or preventing metastasis of the cancer in the subject.

[0010] In a further aspect, the present invention provides a method of increasing survival of a subject suffering from cancer, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer

- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby increasing survival of the subject.

[0011] In another aspect, the present invention further provides use of a radiolabelled agent for binding to CAIX, in the manufacture of a medicament for:

- treating, preventing or minimising progression of cancer in a subject,
- minimising, reducing or preventing growth of a tumour in a subject,
- minimising, reducing or preventing metastasis in a subject, or
- increasing survival of a subject suffering from cancer,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma.

[0012] In another aspect, the present invention further provides use of a radiolabelled agent for binding to CAIX for:

- treating, preventing or minimising progression of cancer in a subject,
- minimising, reducing or preventing growth of a tumor in a subject,
- minimising, reducing or preventing metastasis in a subject, or

- increasing survival of a subject,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma.

[0013] In another aspect, the present invention further provides a radiolabelled agent for binding to CAIX for use in:

- treating, preventing or minimising progression of cancer in a subject,
- minimising, reducing or preventing growth of a tumor in a subject,

- minimising, reducing or preventing metastasis in a subject, or
- increasing survival of a subject.,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma.

[0014] Administration may be by any suitable means, preferably one that allows for systemic administration (eg intravenous infusion) of the agent such that the agent can accumulate at sites in the subject where cell killing is desired, preferably at sites where CAIX is present in or on a tumour. The mode of administration may be dictated by the nature of the agent for binding to CAIX. For example, in the case of an antibody for binding

to CAIX, preferably the agent is administered by intravenous infusion. Labelled peptides or small molecules may be administered orally or by other means.

[0015] In any embodiment, the agent for binding to CAIX may be a small molecule, peptide or polypeptide (such as an antibody or antigen binding fragment thereof).

[0016] In any embodiment, the agent for binding to CAIX is a small molecule, optionally selected from the group consisting of: SLC-0111, SLC-149, SLC-0121, SLC-101, PMI-05, sulfamide-nitroimidazole, JS-403, UB-TT220, HEHEHE-Z09781, -MIP-1486, MIP-1490, MIP-1504 (especially ^{99m}Tc-HEHEHE-Z09781, ^{99m}Tc-MIP-1486, ^{99m}Tc-MIP-1490 or ^{99m}Tc-MIP-1504/5) and PHC-102.

[0017] In any embodiment, the agent for binding to CAIX is a peptide, optionally selected from the group consisting of: 3B-301, 3B-302 or CAIX-P1.

[0018] In any embodiment, the agent for binding to CAIX is a polypeptide, including an antibody or antigen-binding fragment thereof.

[0019] In a particularly preferred embodiments, the agent is in the form of an antibody or antigen binding fragment thereof, most preferably wherein the agent is in the form of an antibody (ie such that the agent at least comprises one or more Fc regions of an antibody).

[0020] In any embodiment, the agent is an antigen binding protein (antibody) such as girentuximab or a functional variant or fragment thereof that retains the ability to bind to CAIX. In some embodiments, the antigen binding protein that binds to or specifically binds to CAIX is G250. In some embodiments, the antigen binding protein that binds to or specifically binds to CAIX is a chimeric antibody or antigen binding fragment thereof. In some embodiments, the antigen binding protein that binds to or specifically binds to CAIX is a humanised antibody or antigen binding fragment thereof. Optionally, the antigen binding protein is humanised G250 (hG250).

[0021] In alternative embodiments, the antibody for binding to CAIX may comprise BCA-356, BAY-794620 or SLC-0131.

[0022] As will be understood, the antibody or fragment thereof, for use according to the invention, is conjugated to a radionuclide. The radionuclide may be conjugated to the antibody directly or indirectly, e.g. by halogenation of amino acid residues. Preferably,

the radionuclide agent is indirectly conjugated to the antibody by way of a linker or chelator moiety.

[0023] In another example, the antibody is conjugated to a bifunctional linker, for example, bromoacetyl, thiols, succinimide ester, TFP ester, a maleimide, or using any amine or thiol- modifying chemistry known in the art.

[0024] The term “chelate” as used herein, refers to an organic compound or portion thereof that can be bonded to a central metal or radiometal atom at two or more points.

[0025] The term “conjugate,” as used herein, refers to a molecule that contains a chelating group or metal complex thereof, a linker group, and which optionally contains a therapeutic moiety, targeting moiety, or cross-linking group.

[0026] Examples of suitable chelating moieties include, but are not limited to, DOTA (1,4,7,10-tetraazacyclododecane-1,4,7,10-tetraacetic acid), DOTMA (1R,4R,7R,10R)- α , α' , α'' , α''' -tetramethyl-1.4.7.10-tetraazacyclododecane-1,4,7,10-tetraacetic acid, DOTAM (1,4,7,10-tetrakis(carbamoylmethyl)-1,4,7,10-tetraazacyclododecane), DOTPA (1,4,7,10-tetraazacyclododecane-1,4,7,10-tetra propionic acid), D03AM-acetic acid (2-(4,7,10-tris(2-amino-2-oxoethyl)-1,4,7,10-tetraazacyclododecan-1-yl)acetic acid), DOTA-GA anhydride (2,2',2''-(10-(2,6-dioxotetrahydro-2H-pyran-3-yl)-1,4,7,10-tetraazacyclododecane-1,4,7-triyl)triacetic acid, DOTP (1,4,7,10-tetraazacyclododecane-1,4,7,10-tetra(methylene phosphonic acid)), DOTMP (1,4,6,10-tetraazacyclododecane-1,4,7,10-tetramethylene phosphonic acid), DOTA-4AMP (1,4,7,10-tetraazacyclododecane-1,4,7,10-tetrakis(acetamido-methylenephosphonic acid), CB-TE2A (1,4,8,11-tetraazabicyclo[6.6.2]hexadecane-4,11-diacetic acid), NOTA (1,4,7-triazacyclononane-1,4,7-triacetic acid), NOTP (1,4,7-triazacyclononane-1,4,7-tri(methylene phosphonic acid), TETPA (1,4,8,11-tetraazacyclotetradecane-1,4,8,11-tetrapropionic acid), TETA (1,4,8,11-tetraazacyclotetradecane-1,4,8,11-tetra acetic acid), HEHA (1,4,7,10,13,16-hexaazacyclohexadecane-1,4,7,10,13,16-hexaacetic acid), PEPA (1,4,7,10,13-pentaazacyclopentadecane-N,N',N'',N''',N''''-pentaacetic acid), H4octapa (N,N'-bis(6-carboxy-2-pyridylmethyl)-ethylenediamine-N,N'-diacetic acid), H2dedpa (1,2-[[6-(carboxy)-pyridin-2-yl]-methylamino]ethane), H6phospa (N,N'-(methylenephosphonate)-N,N'-[6-(methoxycarbonyl)pyridin-2-yl]-methyl-1,2-diaminoethane), TTHA (triethylenetetramine-N,N,N',N'',N''',N''''-hexaacetic acid), DO2P (tetraazacyclododecane dimethanephosphonic acid), HP-DO3A (hydroxypropyltetraazacyclododecanetriacetic

acid), EDTA (ethylenediaminetetraacetic acid), Deferoxamine, DTPA (diethylenetriaminepentaacetic acid), DTPA-BMA (diethylenetriaminepentaacetic acid-bismethylamide), HOPO (octadentate hydroxypyridinones), or porphyrins.

[0027] In some embodiments, radioimmunoconjugates comprise a metal complex of a chelating moiety. For example, chelating groups may be used in metal chelate combinations with metals, such as manganese, iron, and gadolinium and isotopes (e.g., isotopes in the general energy range of 60 to 4,000 keV), such as any of the radioisotopes and radionuclides discussed herein.

[0028] In some embodiments, radioimmunoconjugates comprise a cross-linking group. A cross-linking group is a reactive group that is able to join two or more molecules by a covalent bond. Cross-linking groups may be used to attach the linker and chelating moiety to a therapeutic or targeting moiety. Cross-linking groups may also be used to attach the linker and chelating moiety to a target *in vivo*. In some embodiments, the cross-linking group is an amino-reactive, methionine reactive or thiol-reactive cross-linking group, or a sortase-mediated coupling.

[0029] In some embodiments, the amino-reactive or thiol-reactive cross-linking group comprises an activated ester such as a hydroxysuccinimide ester, 2,3,5,6-tetrafluorophenol ester, 4-nitrophenol ester or an imidate, anhydride, thiol, disulfide, maleimide, azide, alkyne, strained alkyne, strained alkene, halogen, sulfonate, haloacetyl, amine, hydrazide, diazirine, phosphine, tetrazine, isothiocyanate, or oxaziridine. In some embodiments, the sortase recognition sequence may comprise of a terminal glycine-glycine-glycine (GGG) and/or LPTXG amino acid sequence, where X is any amino acid. A person having ordinary skill in the art will understand that the use of cross-linking groups is not limited to the specific constructs disclosed herein, but rather may include other known cross-linking groups.

[0030] Examples of suitable radionuclides for conjugation to an agent for binding to CAIX (such as an antibody for binding to CAIX), and in accordance with the invention, include any radioisotope suitable for treatment of tumour or cancer masses: an alpha emitter, preferably selected from the group consisting of Astatine-211 (^{211}At), Bismuth-212 (^{212}Bi), Bismuth-213 (^{213}Bi), Actinium-225 (^{225}Ac), Radium-223 (^{223}Ra), Lead-212 (^{212}Pb), Thorium-227 (^{227}Th), and Terbium-149 (^{149}Tb), more preferably wherein the radionuclide is Astatine-211 (^{211}At) or Actinium-225 (^{225}Ac).

[0031] In other the radionuclide may be a beta or beta/gamma emitter, preferably selected from the group consisting of: Lutetium-177 (^{177}Lu), Yttrium-90 (^{90}Y), Iodine-131 (^{131}I), Samarium-153 (^{153}Sm), Holmium-166 (^{166}Ho), Rhenium-186 (^{186}Re), or Rhenium-188 (^{188}Re), more preferably wherein the radionuclide is Lutetium-177 (^{177}Lu) or Rhenium-188 (^{188}Re).

[0032] In any embodiment, the agent is an antibody for binding to CAIX (such as girentuximab) and the radioisotope is selected from: actinium-225 (^{225}Ac), astatine-211 (^{211}At), bismuth-212 and bismuth-213 (^{212}Bi , ^{213}Bi), copper-64 and copper-67 (^{64}Cu , ^{67}Cu), iodine -123, -124, -125 or -131 (^{123}I , ^{124}I , ^{125}I , ^{131}I) (^{123}I), lead-212 (^{212}Pb), lutetium-177 (^{177}Lu), radium-223 and radium-224 (^{223}Ra , ^{224}Ra), rhenium-186 and rhenium-188 (^{186}Re and ^{188}Re), samarium-153 (^{153}Sm), and scandium-47 (^{47}Sc), strontium-90 (^{90}Sr), terbium-149 and terbium-161 (^{149}Tb and ^{161}Tb), and yttrium-90 (^{90}Y).

[0033] In particularly preferred embodiments, the agent is an antibody for binding to CAIX (such as girentuximab) and the radioisotope is lutetium-177 (^{177}Lu) or actinium-225 (^{225}Ac), wherein the radioisotope is conjugated to the girentuximab antibody directly or indirectly.

[0034] In any aspect or embodiment, the radioisotope may be conjugated to the girentuximab directly e.g. by halogenation of amino acid residues. Preferably, the radioisotope of the radiolabelled girentuximab is linked indirectly to the girentuximab or antigen binding fragment thereof, for example via a chelator or other linking moiety. In one example, the girentuximab is conjugated to a chelating moiety, selected from the group consisting of: TMT (6,6"-bis[N,N",N""-tetra(carboxymethyl)aminomethyl]-4'-(3-amino-4-methoxyphenyl)-2,2':6',2"-terpyridine), DOTA (1, 4,7,10-tetraazacyclododecane-NN',N"(N""-tetraacetic acid), TCMC, DO3A, CB-DO2A, NOTA, Diamsar, DTPA, CHX-A"-DTPA, TETE, Te2A, HBED, DFO, DFOsq, DFO-NCS and HOPO, a chelator as described in WO 2022/133537 (incorporated herein by reference) or other chelating agent as described herein or known to the skilled person.

[0035] In another example, the radiolabelled girentuximab or antigen binding fragment thereof is conjugated to a bifunctional linker, for example, bromoacetyl, thiols, succinimide ester, TFP ester, a maleimide, or using any amine or thiol- modifying chemistry known in the art.

[0036] In any aspect or embodiment of the invention, it will be understood that the amount of radiolabelled girentuximab or antigen binding fragment thereof administered to a subject, is a therapeutically effective amount of radiolabelled girentuximab or antigen binding fragment thereof.

[0037] In any aspect of the invention, any medicament or pharmaceutical composition described herein may be suitable for administration intraperitoneally, intratumorally, topically, orally, intravenously, to the respiratory tract, preferably by inhalation or intranasally, subcutaneously or intramuscularly. Typically, any medicament or pharmaceutical composition described herein is suitable for administration intravenously.

[0038] In any method or use described herein, the radiolabelled girentuximab or antigen binding fragment thereof may be administered as the sole treatment for a cancer or may be administered in combination with one or more additional treatments. The radiolabelled girentuximab or antigen binding fragment thereof may be administered subsequent to a previous treatment for cancer (optionally wherein the previous treatment comprises radiosensitising treatment, surgery, chemotherapy, immunotherapy, external beam radiation, autologous stem cell therapy, or combinations thereof). The radiolabelled girentuximab or antigen binding fragment thereof may be administered concomitantly with or sequentially with an additional treatment for cancer (optionally wherein the additional treatment comprises surgery, chemotherapy, immunotherapy, external beam radiation, autologous stem cell therapy, or combinations thereof).

[0039] It will be understood that preferably in accordance with the above mentioned aspects and embodiment, that the cancer is preferably one that expresses or overexpresses CAIX.

[0040] In any embodiment, the cancer may be pre-cancerous or non-metastatic. In any embodiment, the cancer may be malignant or metastatic.

[0041] In any embodiment, the methods further comprise identifying a subject in need of treatment as described herein. The method may comprise identifying that the subject has a cancer expressing or overexpressing CAIX.

[0042] In any embodiment, the antibody or antigen binding fragment thereof comprises:

(a) a heavy chain variable domain (VH) comprising three complementarity determining regions (CDRs) of the amino acid sequence as set forth in SEQ ID NOs: 4, 20, 36, 52 or 68; and/or

(b) a light chain variable domain (VL) comprising three complementarity determining regions (CDRs) of the amino acid sequence as set forth in SEQ ID NOs: 84, 100, 116, 132, 148 or 164.

[0043] In any embodiment, the antibody or antigen binding fragment thereof comprises an antigen binding domain that binds specifically to carbonic anhydrase IX (CAIX) and comprises:

FR1 - CDR1 – FR2 – CDR2 – FR3 – CDR3 – FR4 – linker - FR1a - CDR1a – FR2a – CDR2a – FR3a – CDR3a – FR4a

wherein:

FR1, FR2, FR3 and FR4 are each framework regions;

CDR1, CDR2 and CDR3 are each complementarity determining regions;

FR1a, FR2a, FR3a and FR4a are each framework regions;

CDR1a, CDR2a and CDR3a are each complementarity determining regions;

wherein the sequence of any of the complementarity determining regions have an amino acid sequence as described in Table 1 below. Preferably, the framework regions have an amino acid sequence also as described in Table 1 below, including amino acid variation at particular residues which can be determined by aligning the various framework regions derived from each antibody. The CDR1, CDR2 and CDR3 may be sequences from the VH, CDR1a, CDR2a and CDR3a may be sequences from VL, or the CDR1, CDR2 and CDR3 may be sequences from the VL, CDR1a, CDR2a and CDR3a may be sequences from VH.

[0044] In any embodiment, the antigen or antigen binding fragment thereof comprises:

(i) a VH comprising a complementarity determining region (CDR) 1 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least

91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 1, a CDR2 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set in SEQ ID NO: 2, and a CDR3 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 3;

(ii) a VH comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in any of SEQ ID NO: 4, 20, 36, 52 or 68;

(iii) a VL comprising a CDR1 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 81, a CDR2 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 82 and a CDR3 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 83;

(iv) a VL comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%,

at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 84, 100, 116, 132, 148 or 164;

(v) a VH comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 1, a CDR2 comprising a sequence set forth in SEQ ID NO: 2 and a CDR3 comprising a sequence set forth in SEQ ID NO: 3;

(vi) a VH comprising a sequence set forth in any of SEQ ID NO: 4, 20, 36, 52 or 68;

(vii) a VL comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 81, a CDR2 comprising a sequence set forth in SEQ ID NO: 82, and a CDR3 comprising a sequence set forth in SEQ ID NO: 83;

(viii) a VL comprising a sequence set forth in any of SEQ ID NO: 84, 100, 116, 132, 148 or 164;

(ix) a VH comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 1, a CDR2 comprising a sequence set forth between in SEQ ID NO: 2, and a CDR3 comprising a sequence set forth in SEQ ID NO: 3; and a VL comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 81, a CDR2 comprising a sequence set forth in SEQ ID NO: 82, and a CDR3 comprising a sequence set forth in SEQ ID NO: 83; or

(x) a VH comprising a sequence set forth in any of SEQ ID NO: 4, 20, 36, 52 or 68 and a VL comprising a sequence set forth in any of SEQ ID NO: 84, 100, 116, 132, 148 or 164.

[0045] In a further embodiment, the antibody or antigen binding fragment thereof comprises:

(i) a VH comprising a framework region (FR) 1 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 9, 25, 41, 57 or 73; a FR2 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical

to a sequence as set forth in any of SEQ ID Nos: 10, 26, 42, 58 or 74; a FR3 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 11, 27, 43, 59 or 75; a FR4 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 12, 28, 44, 60 or 76; and

(ii) a VL comprising a framework region (FR) 1 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 89, 105, 121, 137, 153 or 169; a FR2 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 90, 106, 122, 138, 154 or 170; a FR3 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 91, 107, 123, 139, 155 or 171; a FR4 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 92, 108, 124, 140, 156 or 172.

[0046] In a further embodiment, the antibody or antigen binding fragment thereof comprises:

(i) a VH comprising a framework region (FR) 1 comprising or consisting of a sequence as set forth in SEQ ID Nos: 9, 25, 41, 57 or 73; a FR2 comprising or consisting of a sequence as set forth in SEQ ID Nos: 10, 26, 42, 58 or 74; a FR3 comprising or consisting of a sequence as set forth in SEQ ID Nos: 11, 27, 43, 59 or 75; a FR4 comprising or consisting of a sequence as set forth in SEQ ID Nos: 12, 28, 44, 60 or 76, and

(ii) a VL comprising a framework region (FR) 1 comprising or consisting of a sequence as set forth in SEQ ID Nos: 89, 105, 121, 137, 153 or 169; a FR2 comprising or consisting of a sequence as set forth in SEQ ID Nos: 90, 106, 122, 138, 154 or 170; a FR3 comprising or consisting of a sequence as set forth in SEQ ID Nos: 91, 107, 123, 139, 155 or 171; a FR4 comprising or consisting of a sequence as set forth in SEQ ID Nos: 92, 108, 124, 140, 156 or 172.

[0047] In any embodiment, the antibody or antigen binding fragment thereof that specifically binds to CAIX comprises an amino acid sequence that consists essentially of or consists of (in order of N to C terminus or C to N terminus) any one of SEQ ID NO: 4, 20, 36, 52 or 68 and/or any one of SEQ ID NO: 84, 100, 116, 132, 148, 164.

[0048] In any embodiment, the antibody or antigen binding fragment thereof comprises:

(a) a heavy chain variable domain (VH) comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in SEQ ID NOs: 4, 20, 36, 52 or 68; and/or

(b) a light chain variable domain (VL) comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in SEQ ID NOs: 84, 100, 116, 132, 148 or 164.

[0049] As described herein, the girentuximab antibody or antigen binding fragment thereof may be in the form of:

- (i) a single domain antibody (sdAb);
- (ii) a single chain Fv fragment (scFv);
- (iii) a dimeric scFv (di-scFv); or
- (iv) one of (ii) or (iii) linked to a constant region of an antibody, Fc or a heavy chain constant domain (CH) 2 and/or CH3.

[0050] Further, as described herein, the girentuximab or antigen binding fragment thereof may be in the form of:

- (i) a diabody;
- (ii) a triabody;
- (iii) a tetrabody;
- (iv) a Fab;
- (v) a F(ab')₂;
- (vi) a Fv;
- (vii) a bispecific antibody or other form of multispecific antibody; or
- (viii) one of (i) to (vii) linked to a constant region of an antibody, Fc or a heavy chain constant domain (CH) 2 and/or CH3.

[0051] Optionally, the variable heavy and variable light regions of the antigen binding domain are joined via a linker.

[0052] In any embodiment, the antigen binding domain may comprise:

FR1 - CDR1 – FR2 – CDR2 – FR3 – CDR3 – FR4 – linker - FR1a - CDR1a – FR2a – CDR2a – FR3a – CDR3a – FR4a.

[0053] As defined herein, the linker may be a chemical, one or more amino acids, or a disulphide bond formed between two cysteine residues.

[0054] Preferably, the girentuximab is in the form of a full IgG antibody.

[0055] In any aspect or embodiment of the present invention, the girentuximab or antigen binding fragment thereof may comprise a human constant region, e.g., an IgG constant region, such as an IgG₁, IgG₂, IgG₃ or IgG₄ constant region or mixtures thereof. In the case of an antibody or protein comprising a V_H and a V_L, the V_H can be linked to a heavy chain constant region and the V_L can be linked to a light chain constant region.

[0056] In one example, the girentuximab or antigen binding fragment thereof comprises a constant region of an IgG₄ antibody or a stabilised constant region of an IgG₄ antibody. In one example, the girentuximab or antigen binding fragment thereof comprises an IgG₄ constant region with a proline at position 241 (according to the numbering system of Kabat (Kabat et al., Sequences of Proteins of Immunological Interest Washington DC United States Department of Health and Human Services, 1987 and/or 1991)).

[0057] In one example, the girentuximab or antigen binding fragment thereof comprises a heavy chain constant region, comprising a stabilised heavy chain constant region, comprising a mixture of sequences fully or partially with or without the C-terminal lysine residue.

[0058] In further embodiments of any aspect herein, the girentuximab or antigen binding fragment thereof comprises an Fc region wherein the Fc region is engineered to have enhanced capacity to induce antibody-dependent cell-mediated cytotoxicity (ADCC). Preferably, the enhanced capacity to induce ADCC is conferred by mutation, deletion or modification of amino acids in the Fc region which interact with an Fc receptor

[0059] In another embodiment, the girentuximab or antigen binding fragment thereof comprises an Fc region that is engineered to:

- have increased *in vitro* or *in vivo* half-life;
- have an increased capacity to induce antibody-dependent cell mediated cytotoxicity (ADCC), antibody-dependent cellular phagocytosis (ADCP) or complement-dependent cytotoxicity; and/or
- have reduced effector function.

[0060] Preferably, the girentuximab antibody is in the form of an antibody (ie comprising a variable light and variable heavy chain, linked to a constant region of an antibody including a heavy chain CH2 and/or CH3).

[0061] In further embodiments, the girentuximab may comprise a heavy chain constant region as defined in SEQ ID NO: 177, 178, 179 or 180 and/or a light chain constant region as defined in SEQ ID NO: 181.

[0062] In certain embodiments, the girentuximab may comprise a heavy chain as set forth in SEQ ID NO: 182, 184, 194, or 185 and/or a light chain as set forth in SEQ ID NO: 186.

[0063] In still further embodiments, the girentuximab is in the form of an antibody and may comprise one or more amino acid substitutions in the constant regions, so as to reduce the *in vivo* half-life of the antibody. The present invention therefore provides for an girentuximab antibody having substitutions in the CH2 and/or CH3 domains of the constant region and comprising substitutions at one or more of residues His310, His435, His436 and Ile253 (Kabat numbering), thereby altering FcRn binding affinity and/or serum half-life of said antibody (eg relative to an girentuximab that comprises a native CH2 and CH3 domain).

[0064] In some examples, the amino acid at position 310 and/or 435 of the antibody may be alanine, glutamic acid, aspartic acid, leucine, isoleucine, arginine, proline, glutamine, methionine, serine, threonine, lysine, asparagine, phenylalanine, tyrosine, tryptophan, cysteine, valine or glycine.

[0065] Preferably, the residue at position 310 is selected from alanine, or glutamic acid or glutamine; or amino acid residue 435 from the heavy chain constant region is selected from arginine, glutamine or alanine. In other preferred embodiments, the antibody has an alanine residue at position 310 and glutamine residue at position 435.

[0066] In a further embodiment, the antibody also comprises an amino acid substitution at residue Lys322. Preferably the substitution is K322A.

[0067] In particularly preferred embodiments, the girentuximab antibody comprises substitutions K322A, H310A and H435Q.

[0068] In a preferred embodiment, the binding affinity for FcRn and/or the serum half-life of the modified antibody is decreased by at least about 2-fold, 3-fold, 4-fold, 5-fold, 10-fold, 15-fold, 20-fold, 25-fold, 30-fold, 40-fold, 50-fold, 60-fold, 70-fold, 80-fold, 90-fold, or 100-fold. In a preferred embodiment of the present invention, the binding affinity for FcRn

and/or the serum half-life of said modified antibody is reduced by at least about 20%, 30%, 40%, 50%, 60%, 70%, 80%, 85%, 90%, 95%, 97%, 98%, or 99%.

[0069] In any aspect or embodiment, the antibody may also comprise amino acid substitutions at residues equivalent to Ser228 and Leu235 of the constant heavy chain region, such as Ser228Pro and/or Leu235Glu.

[0070] As used herein, except where the context requires otherwise, the term "comprise" and variations of the term, such as "comprising", "comprises" and "comprised", are not intended to exclude further additives, components, integers or steps.

[0071] Further aspects of the present invention and further embodiments of the aspects described in the preceding paragraphs will become apparent from the following description, given by way of example and with reference to the accompanying drawings.

Brief description of the drawings

[0072] Figure 1: Determination of binding and biodistribution of ^{177}Lu -DOTA-GmAb. **A.** *In vitro* binding of ^{177}Lu -DOTA-GmAb to various cell lines. **B.** *Ex vivo* biodistribution (ie the distribution of ^{177}Lu -DOTA-GmAb observed in the organs of mice, following necropsy). **C.** Correlation between *in vivo* (ie the distribution of ^{177}Lu -DOTA-GmAb observed in mice following whole-body imaging) and *ex vivo* biodistribution. Closed circles = AsPC-1 radiolabelled-DOTA-GmAb; Closed boxes = FaDu radiolabelled-DOTA-GmAb; Closed triangles = HT-29 radiolabelled-DOTA-GmAb.

[0073] Figure 2: Changes in body weight in mice following injection of ^{177}Lu -DOTA-GmAb. Results are shown mice bearing xenografts derived from AsPc-1 cell line (pancreatic cancer model), FaDu cell line (hypopharyngeal cancer), HT-29 cell line (colorectal cancer).

[0074] Figure 3: Changes in tumour volume in mice following injection of ^{177}Lu -DOTA-GmAb. Results are shown mice bearing xenografts derived from AsPc-1 cell line (pancreatic cancer model), FaDu cell line (hypopharyngeal cancer), HT-29 cell line (colorectal cancer).

[0075] Figure 4: Survival curves from mice following injection of ^{177}Lu -DOTA-GmAb. Results are shown mice bearing xenografts derived from AsPc-1 cell line (pancreatic

cancer model), FaDu cell line (hypopharyngeal cancer), HT-29 cell line (colorectal cancer).

[0076] Figure 5: Imaging of bladder cancer. Day 0: whole body ^{89}Zr -girentuximab scans: **A:** Coronal PET/CT fusion. **B:** maximum intensity projection (MIP) visualisation.

[0077] Figure 6: Imaging of bladder cancer. Day 2, **A.** ^{89}Zr -girentuximab pelvis PET/CT fusion images: (arrows) radiopharmaceutical uptake on different sides of bladder wall. **B.** 3D representation with superimposed image of bladder.

Detailed description of the embodiments

[0078] The present invention relates to the treatment of cancers for which there is currently a lack of robust oncology management options. To improve the morbidity and mortality of these oncologic indications, therapeutic innovation is needed.

[0079] The use of radiolabelled CAIX-binding agent is known in the context of treatment of renal cell carcinomas. However, prior to the present invention, it was not known whether the molecular targeting of CAIX could be used to successfully treat other solid cancers which may express CAIX.

[0080] Although CAIX is typically associated with advanced disease, it is not known whether molecular targeting of CAIX for radioimmunotherapy is possible during the early stages of certain cancers, or if the cancer can only be targeted in late disease. Further, some cancer exhibit reduced expression as disease progresses and it is therefore also not clear whether CAIX targeting is a useful means for treating these types of cancers.

[0081] Moreover, given the heterogeneity of many cancer, the simple presence of expression of CAIX (eg as determined by immunohistochemistry techniques), does not necessarily indicate that the cancer could be treated using molecular targeting radioimmunotherapeutic methods. For example, in a recent study published by Huizing et al (2021, *Physics and Imaging in Radiation Oncology*, 145-150), it was found that a ^{111}In -labelled F(ab')₂ form of the CAIX-binding antibody girentuxumab, was not able to discriminate between tumour and non-tumour cells and it was therefore concluded that this agent would not be useful for imaging of head and neck cancers. In contrast, the present inventors have demonstrated that radiolabelled girentuximab is useful for the

treatment of the same cancer cell type; demonstrating that the outcomes in diagnostic studies are not necessarily predictive of the outcome in therapeutic studies.

[0082] The present invention is therefore based on the finding that a subset of cancers having CAIX expression can indeed be successfully treated using a radiolabelled agent for binding to CAIX.

General definitions

[0083] Throughout this specification, unless specifically stated otherwise or the context requires otherwise, reference to a single step, composition of matter, group of steps or group of compositions of matter shall be taken to encompass one and a plurality (i.e. one or more) of those steps, compositions of matter, groups of steps or groups of compositions of matter. Thus, as used herein, the singular forms “a”, “an” and “the” include plural aspects, and vice versa, unless the context clearly dictates otherwise. For example, reference to “a” includes a single as well as two or more; reference to “an” includes a single as well as two or more; reference to “the” includes a single as well as two or more and so forth.

[0084] Those skilled in the art will appreciate that the present invention is susceptible to variations and modifications other than those specifically described. It is to be understood that the invention includes all such variations and modifications. The invention also includes all of the steps, features, compositions and compounds referred to or indicated in this specification, individually or collectively, and any and all combinations or any two or more of said steps or features.

[0085] One skilled in the art will recognize many methods and materials similar or equivalent to those described herein, which could be used in the practice of the present invention. The present invention is in no way limited to the methods and materials described.

[0086] All of the patents and publications referred to herein are incorporated by reference in their entirety.

[0087] The present invention is not to be limited in scope by the specific examples described herein, which are intended for the purpose of exemplification only. Functionally-

equivalent products, compositions and methods are clearly within the scope of the present invention.

[0088] Any example or embodiment of the present invention herein shall be taken to apply mutatis mutandis to any other example or embodiment of the invention unless specifically stated otherwise.

[0089] Unless specifically defined otherwise, all technical and scientific terms used herein shall be taken to have the same meaning as commonly understood by one of ordinary skill in the art (for example, in diagnostic technology, radioimaging, cell culture, molecular genetics, immunology, immunohistochemistry, protein chemistry, and biochemistry).

[0090] The term “and/or”, e.g., “X and/or Y” shall be understood to mean either “X and Y” or “X or Y” and shall be taken to provide explicit support for both meanings or for either meaning.

Carbonic anhydrase IX

[0091] As used herein, carbonic anhydrase is also known as: CA-IX, CA9, CAIX, Carbonate dehydratase IX, Carbonic anhydrase 9, Carbonic anhydrase IX, carbonic dehydratase, G250, Membrane antigen MN, P54/58N, pMW1, RCC-associated antigen G250, RCC-associated protein G250, and Renal cell carcinoma-associated antigen G250.

[0092] Cancer cells primarily express the plasma-membrane-associated CA isoforms CAIX and CAXII, as well as intracellular CAs such as CAI and CAII. Amongst the cancer-related CAs, CAIX has gained most attention, since expression of this isoform in healthy tissue is restricted to epithelial cells in the stomach and gut, but is strongly upregulated in renal cancers.

[0093] CAIX, the expression of which is under control of the hypoxia-inducible factor 1 (HIF-1), is predominantly located in chronically hypoxic tumour regions. However, CAIX can also be found in mild hypoxic or even normoxic regions, since the expression of CAIX can be activated by components of the mitogen-activated protein kinase (MAPK) pathway.

[0094] The agent for binding to CAIX and for use in accordance with the methods of the invention, may be any compound that specifically recognises or binds to CAIX, mediates its activity by binding to CAIX or a fragment or splice variant thereof; irreversibly binds at the entrance to the active site, and/or inhibits CAIX by coordinating to the zinc ion at the active site of CAIX.

[0095] Preferably the agent for binding to CAIX specifically interacts with a CAIX polypeptide. Specifically interacting with (e.g. recognising or binding to) means that the agent e.g. antibody, has a greater affinity for CAIX compared to other polypeptides. In one embodiment the agent interacts with (i.e. binds to or recognises) or modulates the activity of CAIX polypeptide and/or mediates an antibody dependent cell cytotoxicity (ADCC) and/or complement mediated cytotoxicity (CDC). Thus, according to one embodiment, the agent is a CAIX inhibitor. Said CAIX inhibitor may act on the protein level or the nucleic acid level. Examples for CAIX inhibitors e.g. acting on the protein level include but are not limited to peptides and anti-CAIX antibodies as well as functional fragments of those antibodies or small organic molecules, preferably having a molecular weight below 500 g/mol.

[0096] Examples of anti-CAIX antibodies or antibodies for binding to CAIX are described in EP 637 336, WO 93/18152, WO 95/34650, WO 00/24913, WO 02/063010, WO 04/025302, WO 05/037083, WO 201 1/139375, Murri-Plesko et al., Eur J Pharmacol 201 1 , 657: 173-183.

[0097] Examples of small organic molecules for binding to CAIX include but are not limited to sulphonamides, heteroaromatic sulphonamides, sulfamates, coumarins and thiocoumarins and BAY-79-4620. Examples for inhibitors acting on the nucleic acid level are siRNA molecules, ribozymes and/or antisense molecules.

[0098] As used herein, the term “specifically binds” or “binds specifically” shall be taken to mean that an agent for use according to the invention, reacts or associates more frequently, more rapidly, with greater duration and/or with greater affinity with a CAIX or cell expressing same than it does with alternative antigens or cells. For example, an antigen binding protein that binds to CAIX with materially greater affinity (e.g., 1.5 fold or 2 fold or 5 fold or 10 fold or 20 fold or 40 fold or 60 fold or 80 fold to 100 fold or 150 fold or 200 fold) than it does to other antigens.

[0099] Methods for assessing binding to a protein (eg CAIX) are known in the art, e.g., as described in Scopes (In: Protein purification: principles and practice, Third Edition, Springer Verlag, 1994). Such a method generally involves immobilizing the agent (eg antibody) and contacting it with labeled target (in the case of an antibody, the antigen). Following washing to remove non-specific bound protein, the amount of label and, as a consequence, bound antigen is detected. Of course, the antigen binding site can be labeled and the antigen immobilized. Panning-type assays can also be used. Alternatively, or additionally, surface plasmon resonance assays can be used.

[0100] Other standard methods for assessing binding of to a target, such as CAIX, are also known in the art.

Small molecules

[0101] In any embodiment, the agent for binding to CAIX is the small molecule SLC-0111 (CAS 178606-66-1), SLC-149 (as described in EP 3317255 B1, incorporated herein by reference), SLC-0121 or SLC-101.

[0102] In any embodiment, the agent for binding to CAIX is the small molecule/contrast dye PMI-05 (as described in US2019/0192699A1, incorporated herein by reference).

[0103] In any embodiment, the agent for binding to CAIX is the small molecule sulfamide-nitroimidazole (as described in Rami et al., (2013), J. Med. Chem, 56: 8512-8520, incorporated herein by reference).

[0104] In any embodiment, the agent for binding to CAIX is the small molecule JS-403 (as described in WO 2010/089752 A1, incorporated herein by reference).

[0105] In any embodiment, the agent for binding to CAIX is the small molecule UB-TT220 (as described in WO 2022/015955 A1, incorporated herein by reference).

[0106] In any embodiment, the agent for binding to CAIX is the small molecule ^{99m}Tc-HEHEHE-Z09781 (Kim et al., (2017) Advanced Science, 4:1600471; Gebauer and Skerra (2009) Current Opin in Chem Biol, 13(3):245-55; Schardt et al., (2017) Mol Pharmaceutics, 14(4):1047-56; Tolmachev et al., (2008) Bioconjugate Chem, 19(8):1579-87; Liu et al., (2022) Analytical and Bioanalytical Chemistry, 414:1095-1104; Grindel et al., (2022) ACS Chem Biol, 17(6): 1543-55, incorporated herein by reference), ^{99m}Tc-MIP-1486, ^{99m}Tc-MIP-1490 (4-(2-bis((1-(2-((1,5-dicarboxy-3-(2-carboxyethyl)pentan-3-

yl)amino)-2-oxoethyl)-1H-imidazol-2-yl)methyl)amino-X)benzenesulfonamide where X= ethyl) or ^{99m}Tc-MIP-1504 (4-(2-bis((1-(2-((1,5-dicarboxy-3-(2-carboxyethyl)pentan-3-yl)amino)-2-oxoethyl)-1H-imidazol-2-yl)methyl)amino-X)benzenesulfonamide where X= n-butyloxy) (Hillier et al., (2012) Journal of Nuclear Medicine, 53(s1):217, incorporated herein by reference)

[0107] In any embodiment, the agent for binding to CAIX is the small molecule PHC-102 (as described in WO 2015/114171 A1; WO 2018154517 A1; US 2014/0357650 A1; WO 2015/114171 A1, incorporated herein by reference)

Peptides

[0108] In any embodiment, the agent for binding to CAIX is a peptide. As used herein, a peptide will be understood to comprise a chain of more than 1 amino acid residues. Typically, a peptide may comprise from about 2 to 30 amino acids, e.g. from about 5 to 30, from about 10 to 30, from about 2 to 25, from about 5 to 25, from about 10 to 25, or from about 10 to 20 amino acids. A peptide may have a length of at least 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 or 30 amino acids. Typically a peptide is no longer than about 40 amino acids, e.g. no longer than about 35, 30, 25, 20, 17, 15, 14, 13, 12, 11 or 10 amino acids.

[0109] In any embodiment, the agent for binding to CAIX is the peptide 3B-301 (also known as Debio 0228; Queen et al., (2018) Int J of Biol Macromol, 106:840-850; Eldehna et al., (2019) Bioorganic Chem, 90:103102; Lavecchia et al., (2011) Carbohydrate Res, 346(3):442-48; Krymov et al., (2022) Eur J of Medicinal Chem, 228:113997; Supuran (2008) BJI Int, 101(s4):39-40; Koyuncu et al., (2019) J of Enzyme Inhibition and Medicinal Chem, 34(1):703-11; Kumar et al., (2017) Eur J of Medicinal Chem, 136:52-62, incorporated herein by reference) or 3B-302.

[0110] In any embodiment, the agent for binding to CAIX is the peptide CAIX-P1, having the amino acid sequence YNTNHVPLSPKY (as described in Askoxylakis et al., (2010), PLoS One, 5(12): e15962), optionally wherein the peptide is labelled with ¹²⁵I or ¹³¹I, for enabling detection thereof (although it will be appreciated that any suitable radiolabel or other detectable moiety may be used).

Polypeptides and antibodies

[0111] According to especially preferred embodiments, the agent comprises an anti-CAIX antibody and/or a functional fragment of such an antibody. The fragment of the anti-CAIX antibody may have essentially the same CAIX-binding and/or inhibiting activity as the full-length anti-CAIX antibody and/or is an epitope-binding fragment of the anti-CAIX antibody.

[0112] Reference herein to an antibody or antigen binding fragment thereof that “binds to” carbonic anhydrase IX (CAIX) provides literal support for an antibody or fragment thereof that “binds specifically to” or “specifically binds to” CAIX.

[0113] The antibody and/or the antibody fragment thereof may be selected from the group consisting of polyclonal antibodies, monoclonal antibodies, antigen-binding fragments thereof such as F(ab')₂, Fab', scFv, dsFv and chimerized, humanized and fully human variants thereof. The antibody may be multivalent, or multivalent and multispecific.

[0114] In particularly preferred embodiments, the antibody is a full antibody, comprising an antigen binding domain (Fab) of an antibody, and at least one Fc region of an antibody. The antibody may include human constant regions of IgG1, IgG2a, IgG3, or IgG4.

[0115] According to a further preferred embodiment, an anti-CAIX antibody or epitope-binding fragment thereof for use according to the invention, binds to the amino acid sequence LSTAFARV and/or ALGPGREYRAL.

[0116] In any embodiment, the agent for binding to CAIX is in the form of the antibody BAY-794620, or an antigen binding fragment thereof (as described in WO 2003/100029 A2; WO 2003/033674 A2; Theiner et al., (2021) Tierarztl Prax Ausg G Grosstiere Nutztiere, 49(6): 392-402; Kimani et al., (2011) Photochemistry and Photobiology, 88(1):175-87; NCT01065623 (v24, 30 September 2014); NCT01028755 (v30, 19 January 2015), incorporated herein by reference).

[0117] In any embodiment, the agent for binding to CAIX is in the form of the antibody SLC-0131, or an antigen binding fragment thereof.

[0118] According to further particularly preferred embodiments, the agent for binding to CAIX is the antibody anti-G250 (sometimes also called G250 antibody) and/or an antigen-binding fragment thereof. Anti-G250 antibodies are, e.g., described in EP-B-0

637 336. The antibody or fragment thereof may be chimeric or humanised G250 antibody. In some embodiments, the antigen binding protein that binds to or specifically binds to CAIX is as described in any of WO 2002/062972 A2 (US 2004/0219633 A1), WO 2004/002526 A1 (US 7,632,496 B2), WO 2006/002889 A2 (US 7,691,375 B2), WO 2009/056342 A1 (US 2014/0017252 A1), WO 2011/032973 A1 (US 2012/0207672 A1), and WO 2014/128258 A1 (US 10,620,208 B2), or WO 2021/000017 A1, the entire contents of each of these publications is incorporated herein by reference.

[0119] The antibodies for use in the present invention may be produced by any suitable method known in the art including but not limited by methods as described in PCT/EP02/01282 and PCT/EP02/01283, which are incorporated herein by reference.

[0120] An especially preferred antibody is cG250, preferably girentuximab (INN). Another especially preferred embodiment is the monoclonal antibody G250 produced by the hybridoma cell line DSM ACC 2526. The antibody cG250 is an IgG1 kappa light chain chimeric version of an originally murine monoclonal antibody mG250.

[0121] Variants of the original chimeric G250 (cG250) antibody are known, including WX-G250 and WX-G250RIT (131 iodine) (Janssen Global Services LLC).

[0122] In a particularly preferred embodiment, the antibody is ¹⁷⁷Lu-girentuximab.

[0123] In any embodiment, the antibody or antigen binding fragment thereof comprises:

(a) a heavy chain variable domain (VH) comprising three complementarity determining regions (CDRs) of the amino acid sequence as set forth in any of SEQ ID NOs: 4, 20, 36, 52 or 68; and/or

(b) a light chain variable domain (VL) comprising three complementarity determining regions (CDRs) of the amino acid sequence as set forth in any of SEQ ID NOs: 84, 100, 116, 132, 148 or 164.

[0124] In any embodiment, the antibody or antigen binding fragment thereof comprises an antigen binding domain that binds specifically to carbonic anhydrase IX (CAIX) and comprises:

FR1 - CDR1 – FR2 – CDR2 – FR3 – CDR3 – FR4 – linker - FR1a - CDR1a – FR2a – CDR2a – FR3a – CDR3a – FR4a

wherein:

FR1, FR2, FR3 and FR4 are each framework regions;

CDR1, CDR2 and CDR3 are each complementarity determining regions;

FR1a, FR2a, FR3a and FR4a are each framework regions;

CDR1a, CDR2a and CDR3a are each complementarity determining regions;

wherein the sequence of any of the complementarity determining regions have an amino acid sequence as described in Table 1 below. Preferably, the framework regions have an amino acid sequence also as described in Table 1 below, including amino acid variation at particular residues which can be determined by aligning the various framework regions derived from each antibody. The CDR1, CDR2 and CDR3 may be sequences from the VH, CDR1a, CDR2a and CDR3a may be sequences from VL, or the CDR1, CDR2 and CDR3 may be sequences from the VL, CDR1a, CDR2a and CDR3a may be sequences from VH.

[0125] In any embodiment, the antigen or antigen binding fragment thereof comprises:

(i) a VH comprising a complementarity determining region (CDR) 1 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 1, a CDR2 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set in SEQ ID NO: 2, and a CDR3 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 3;

(ii) a VH comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least

89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in any of SEQ ID NO: 4, 20, 36, 52 or 68;

(iii) a VL comprising a CDR1 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 81, a CDR2 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 82 and a CDR3 comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 83;

(iv) a VL comprising a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% or at least 99% identical to a sequence set forth in SEQ ID NO: 84, 100, 116, 132, 148 or 164;

(v) a VH comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 1, a CDR2 comprising a sequence set forth in SEQ ID NO: 2 and a CDR3 comprising a sequence set forth in SEQ ID NO: 3;

(vi) a VH comprising a sequence set forth in any of SEQ ID NO: 4, 20, 36, 52 or 68;

(vii) a VL comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 81, a CDR2 comprising a sequence set forth in SEQ ID NO: 82, and a CDR3 comprising a sequence set forth in SEQ ID NO: 83;

(viii) a VL comprising a sequence set forth in any of SEQ ID NO: 84, 100, 116, 132, 148 or 164;

(ix) a VH comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 1, a CDR2 comprising a sequence set forth between in SEQ ID NO: 2, and a CDR3 comprising a sequence set forth in SEQ ID NO: 3; and a VL comprising a CDR1 comprising a sequence set forth in SEQ ID NO: 81, a CDR2 comprising a sequence set forth in SEQ ID NO: 82, and a CDR3 comprising a sequence set forth in SEQ ID NO: 83; or

(x) a VH comprising a sequence set forth in any of SEQ ID NO: 4, 20, 36, 52 or 68 and a VL comprising a sequence set forth in any of SEQ ID NO: 84, 100, 116, 132, 148 or 164.

[0126] In a further embodiment, the antibody or antigen binding fragment thereof comprises:

(i) a VH comprising a framework region (FR) 1 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 9, 25, 41, 57 or 73; a FR2 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 10, 26, 42, 58 or 74; a FR3 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 11, 27, 43, 59 or 75; a FR4 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 12, 28, 44, 60 or 76; and

(ii) a VL comprising a framework region (FR) 1 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least

91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 89, 105, 121, 137, 153 or 169; a FR2 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 90, 106, 122, 138, 154 or 170; a FR3 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 91, 107, 123, 139, 155 or 171; a FR4 comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in any of SEQ ID Nos: 92, 108, 124, 140, 156 or 172.

[0127] In a further embodiment, the antibody or antigen binding fragment thereof comprises:

(i) a VH comprising a framework region (FR) 1 comprising or consisting of a sequence as set forth in SEQ ID Nos: 9, 25, 41, 57 or 73; a FR2 comprising or consisting of a sequence as set forth in SEQ ID Nos: 10, 26, 42, 58 or 74; a FR3 comprising or consisting of a sequence as set forth in SEQ ID Nos: 11, 27, 43, 59 or 75; a FR4 comprising or consisting of a sequence as set forth in SEQ ID Nos: 12, 28, 44, 60 or 76, and

(ii) a VL comprising a framework region (FR) 1 comprising or consisting of a sequence as set forth in SEQ ID Nos: 89, 105, 121, 137, 153 or 169; a FR2 comprising or consisting of a sequence as set forth in SEQ ID Nos: 90, 106, 122, 138, 154 or 170; a FR3 comprising or consisting of a sequence as set forth in SEQ ID Nos: 91, 107, 123, 139, 155 or 171; a FR4 comprising or consisting of a sequence as set forth in SEQ ID Nos: 92, 108, 124, 140, 156 or 172.

[0128] In any embodiment, the antibody or antigen binding fragment thereof that specifically binds to CAIX comprises an amino acid sequence that consists essentially of or consists of (in order of N to C terminus or C to N terminus) any one of SEQ ID NO: 4, 20, 36, 52 or 68 and/or any one of SEQ ID NO: 84, 100, 116, 132, 148, 164.

[0129] In any embodiment, the antibody or antigen binding fragment thereof comprises:

(a) a heavy chain variable domain (VH) comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in SEQ ID NOs: 4, 20, 36, 52 or 68; and/or

(b) a light chain variable domain (VL) comprising or consisting of a sequence at least about 80%, at least 81%, at least 82%, at least 83%, at least 84%, at least 85%, at least 86%, at least 87%, at least 88%, at least 89%, at least 90%, at least 91%, at least 92%, at least 93%, at least 94%, at least 95%, at least 96%, at least 97%, at least 98% at least 99% identical to a sequence as set forth in SEQ ID NOs: 84, 100, 116, 132, 148 or 164.

[0130] In any embodiment, the antibody or antigen binding fragment thereof for binding to CAIX may be in the form of:

- (i) a single chain Fv fragment (scFv);
- (ii) a dimeric scFv (di-scFv); or
- (iii) one of (i) or (ii) linked to a constant region of an antibody, Fc or a heavy chain constant domain (CH) 2 and/or CH3.

[0131] In any embodiment, the antibody or antigen binding fragment thereof for binding to CAIX may be in the form of:

- (i) a diabody;
- (ii) a triabody;
- (iii) a tetrabody;

- (iv) a Fab;
- (v) a F(ab')₂;
- (vi) a Fv; or
- (vii) one of (i) to (vi) linked to a constant region of an antibody, Fc or a heavy chain constant domain (CH) 2 and/or CH3.

[0132] In any embodiment, the antibody or antigen binding fragment thereof for use according to the invention may be a fusion protein comprising an antigen binding protein, immunoglobulin variable domain, antibody, dab (single domain antibody), di-scFv, scFv, Fab, Fab', F(ab')₂, Fv fragment, diabody, triabody, tetrabody, linear antibody, single-chain antibody molecule, or multispecific antibody as described herein.

[0133] An antigen binding fragment, immunoglobulin variable domain, antibody, dab, di-scFv, scFv, Fab, Fab', F(ab')₂, Fv fragment, diabody, triabody, tetrabody, linear antibody, single-chain antibody molecule, or multispecific antibody, fusion protein or conjugate as described herein may be obtained by expressing a nucleic acid encoding the same.

[0134] An antibody or antigen binding fragment thereof as described herein may comprise a human constant region, e.g., an IgG constant region, such as an IgG1, IgG2, IgG3 or IgG4 constant region or mixtures thereof. In the case of an antibody or protein comprising a VH and a VL, the VH can be linked to a heavy chain constant region and the VL can be linked to a light chain constant region.

[0135] In one example an antibody or antigen binding fragment thereof as described herein comprises a heavy chain constant region, comprising a stabilized heavy chain constant region, comprising a mixture of sequences fully or partially with or without the C-terminal lysine residue.

[0136] In one example, an antibody or antigen binding fragment thereof as described herein comprises a VH disclosed herein linked or fused to an IgG4 constant region or stabilized IgG4 constant region (e.g., as discussed above) and the VL is linked to or fused to a kappa light chain constant region.

[0137] The functional characteristics of an antigen binding fragment thereof as described herein will be taken to apply *mutatis mutandis* to an antibody as described herein.

[0138] An antibody or antigen binding fragment thereof for a use as described herein may be purified, substantially purified, isolated and/or recombinant.

Table 1: Summary of amino acid and nucleotide sequences of preferred CAIX-binding antibodies

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
Girentuximab chimeric VHO Variable Heavy chain	HCDR1 (protein)	1	NYYMS
	HCDR2 (protein)	2	AINSDGGITYYLDTVKG
	HCDR3 (protein)	3	HRSGYFSMDY
	VH (protein)	4	DVKLVESGGGLVKLGSLKLSAASGFTFSNYYMSWV RQTPEKRLELVA AINSDGGITYYLDTVKGRFTISRDNK NTLYLQMSSLKSEDTALFYCARHRSGYFSMDYWGQG TSVTVSS
	HCDR1 (DNA)	5	AACTATTACATGTCT
	HCDR2 (DNA)	6	GCCATTAATAGTGATGGTGGTATCACCTACTATCTAG AACTGTGAAGGGC
	HCDR3 (DNA)	7	CACCGCTCGGGCTACTTTTCTATGGACTAC
	VH (DNA)	8	GACGTGAAGCTCGTGGAGTCTGGGGGAGGCTTAGT GAAGCTTGGAGGATCCCTGAACTCTCCTGTGCAGC CTCTGGATTCACTTTCAGTAACTATTACATGTCTTGG GTTCCGCCAGACTCCAGAGAAGAGGCTGGAGTTGGT CGCAGCCATTAATAGTGATGGTGGTATCACCTACTA TCTAGACACTGTGAAGGGCCGATTCAACATTTCAAG AGACAATGCCAAGAACACCCTGTACCTGCAAATGAG CAGTCTGAAGTCTGAGGACACAGCCTTGTTTTACTG TGCAAGACACCGCTCGGGCTACTTTTCTATGGACTA CTGGGGTCAAGGAACCTCAGTCACCGTCTCCTCA
	HFR1 (protein)	9	DVKLVESGGGLVKLGSLKLSAASGFTFS
	HFR2 (protein)	10	WVRQTPEKRLELVA
	HFR3 (protein)	11	RFTISRDNKNTLYLQMSSLKSEDTALFYCAR
	HFR4 (protein)	12	WGQGTSTVTSS
	HFR1 (DNA)	13	GACGTGAAGCTCGTGGAGTCTGGGGGAGGCTTAGT GAAGCTTGGAGGATCCCTGAACTCTCCTGTGCAGC CTCTGGATTCACTTTCAGT

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	HFR2 (DNA)	14	TGGGTTCCGCCAGACTCCAGAGAAGAGGCTGGAGTTGGTCGCA
	HFR3 (DNA)	15	CGATTCACCATTTCAGAGACAATGCCAAGAACACCCTGTACCTGCAAATGAGCAGTCTGAAGTCTGAGGACACAGCCTTGTTTTACTGTGCAAGA
	HFR4 (DNA)	16	TGGGGTCAAGGAACCTCAGTCACCGTCTCCTCA
Girentuximab humanised VH1 Variable Heavy chain	HCDR1 (protein)	17	NYYSMS
	HCDR2 (protein)	18	AINSDGGITYYLDTVKG
	HCDR3 (protein)	19	HRSGYFSMDY
	VH (protein)	20	DVKLVESGGGLVKPGGSLRLSCAASGFTFSNYYSMSWVRQAPGKGLVLA AINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMSSLKSEDTALYYCARHRSGYFSMDYWGQGTSVTVSS
	HCDR1 (DNA)	21	AACTACTACATGAGC
	HCDR2 (DNA)	22	GCCATTAAACAGTGACGGTGGCATCACCTACTACCTG GACACCGTGAAGGGC
	HCDR3 (DNA)	23	CACAGGAGCGGCTACTTCTCTATGGACTAC
	VH (DNA)	24	GACGTGAAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGTAACTACTACATGAGCT GGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGCT GGTGCGCCGCAATTAACAGTGACGGTGGCATCACCTA CTACCTGGACACCGTGAAGGGCCGATTACCATCTC CAGGGACAACGCCAAGAACACCCTGTATCTGCAAAT GAGCAGCCTGAAGAGCGAGGACACGGCCCTGTATT ACTGTGCGAGACACAGGAGCGGCTACTTCTCTATGG ACTACTGGGGCCAGGGCACCAGCGTCACTGTCTCC TCA
	HFR1 (protein)	25	DVKLVESGGGLVKPGGSLRLSCAASGFTFS
	HFR2 (protein)	26	WVRQAPGKGLVLA
	HFR3 (protein)	27	RFTISRDN AKNTLYLQMSSLKSEDTALYYCAR
	HFR4 (protein)	28	WGQGTSVTVSS
	HFR1 (DNA)	29	GACGTGAAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGT
	HFR2 (DNA)	30	TGGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGC TGGTTGCC

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	HFR3 (DNA)	31	CGATTCACCATCTCCAGGGACAACGCCAAGAACACC CTGTATCTGCAAATGAGCAGCCTGAAGAGCGAGGA CACGGCCCTGTATTACTGTGCGAGA
	HFR4 (DNA)	32	TGGGGCCAGGGCACCAGCGTCACTGTCTCCTCA
Girentuximab humanised VH3 Variable Heavy chain	HCDR1 (protein)	33	NYYSMS
	HCDR2 (protein)	34	AINSDGGITYYLDTVKG
	HCDR3 (protein)	35	HRSGYFSMDY
	VH (protein)	36	DVKLVESGGGLVKPGGSLRLSCAASGFTFSNYMSW VRQAPGKLELVAAINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMNSLRAEDTALYYCARHRSGYFSMDYWG QGTLTVSS
	HCDR1 (DNA)	37	AACTACTACATGAGC
	HCDR2 (DNA)	38	CCATTAACAGTGACGGTGGCATCACCTACTACCTGG ACACCGTGAAGGGC
	HCDR3 (DNA)	39	CACAGGAGCGGCTACTTCTCTATGGACTAC
	VH (DNA)	40	GACGTGAAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGTAACTACTACATGAGCT GGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGCT GGTTGCCGCCATTAACAGTGACGGTGGCATCACCTA CTACCTGGACACCGTGAAGGGCCGATTACCATCTC CAGGGACAACGCCAAGAACACCCTGTATCTGCAAAT GAACAGCCTGAGGGCCGAGGACACGGCCCTGTATT ACTGTGCGAGACACAGGAGCGGCTACTTCTCTATGG ACTACTGGGGCCAGGGCACCCCTGGTCACTGTCTCC TCA
	HFR1 (protein)	41	DVKLVESGGGLVKPGGSLRLSCAASGFTFS
	HFR2 (protein)	42	WVRQAPGKLELVA
	HFR3 (protein)	43	RFTISRDNKNTLYLQMNSLRAEDTALYYCAR
	HFR4 (protein)	44	WGQGLTVTVSS
	HFR1 (DNA)	45	GACGTGAAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGT
	HFR2 (DNA)	46	TGGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGC TGTTGCC
	HFR3 (DNA)	47	CGATTCACCATCTCCAGGGACAACGCCAAGAACACC CTGTATCTGCAAATGAACAGCCTGAGGGCCGAGGA CACGGCCCTGTATTACTGTGCGAGA

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	HFR4 (DNA)	48	TGGGGCCAGGGCACCCCTGGTCACTGTCTCCTCA
Girentuximab	HCDR1 (protein)	49	NYYMS
humanised VH4	HCDR2 (protein)	50	AINSDGGITYYLDTVKG
Variable Heavy chain	HCDR3 (protein)	51	HRSGYFSMDY
	VH (protein)	52	EVQLVESGGGLVKPGGSLRLSCAASGFTFSNYYMSW VRQAPGKGLELVAAINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMNSLRAEDTALYYCARHRSGYFSMDYWG QGTLTVSS
	HCDR1 (DNA)	53	AACTACTACATGAGC
	HCDR2 (DNA)	54	GCCATTAAACAGTGACGGTGGCATCACCTACTACCTG GACACCGTGAAGGGC
	HCDR3 (DNA)	55	CACAGGAGCGGCTACTTCTCTATGGACTAC
	VH (DNA)	56	GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGTAACTACTACATGAGCT GGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGCT GGTTGCCGCCATTAACAGTGACGGTGGCATCACCTA CTACCTGGACACCGTGAAGGGCCGATTACCATCTC CAGGGACAACGCCAAGAACACCCTGTATCTGCAAAT GAACAGCCTGAGGGCCGAGGACACGGCCCTGTATT ACTGTGCGAGACACAGGAGCGGCTACTTCTCTATGG ACTACTGGGGCCAGGGCACCCCTGGTCACTGTCTCC TCA
	HFR1 (protein)	57	EVQLVESGGGLVKPGGSLRLSCAASGFTFS
	HFR2 (protein)	58	WVRQAPGKGLELVA
	HFR3 (protein)	59	RFTISRDNKNTLYLQMNSLRAEDTALYYCAR
	HFR4 (protein)	60	WGQGTLTVSS
	HFR1 (DNA)	61	GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGT
	HFR2 (DNA)	62	TGGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGC TGGTTGCC
	HFR3 (DNA)	63	CGATTACCATCTCCAGGGACAACGCCAAGAACACC CTGTATCTGCAAATGAACAGCCTGAGGGCCGAGGA CACGGCCCTGTATTACTGTGCGAGA
	HFR4 (DNA)	64	TGGGGCCAGGGCACCCCTGGTCACTGTCTCCTCA
Girentuximab humanised VH5	HCDR1 (protein)	65	NYYMS

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
Variable Heavy chain	HCDR2 (protein)	66	AINSDGGITYYLDTVKG
	HCDR3 (protein)	67	HRSGYFSMDY
	VH (protein)	68	EVQLVESGGGLVKPGGSLRLSCAASGFTFSNYYMSW VRQAPGKGLEWVA AINSDGGITYYLDTVKG RFTISRDN AKNTLYLQMNSLRAEDTALYYCAR HRSGYFSMDYWG QGTLTVSS
	HCDR1 (DNA)	69	AACTACTACATGAGC
	HCDR2 (DNA)	70	GCCATTAACAGTGACGGTGGCATCACCTACTACCTG GACACCGTGAAGGGC
	HCDR3 (DNA)	71	CACAGGAGCGGCTACTTCTCTATGGACTAC
	VH (DNA)	72	GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGTAACTACTACATGAGCT GGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGTG GGTTGCCGCCATTAACAGTGACGGTGGCATCACCTA CTACCTGGACACCGTGAAGGGCCGATTACCATCTC CAGGGACAACGCCAAGAACACCCTGTATCTGCAAT GAACAGCCTGAGGGCCGAGGACACGGCCCTGTATT ACTGTGCGAGACACAGGAGCGGCTACTTCTCTATGG ACTACTGGGGCCAGGGCACCCCTGGTCACTGTCTCC TCA
	HFR1 (protein)	73	EVQLVESGGGLVKPGGSLRLSCAASGFTFS
	HFR2 (protein)	74	WVRQAPGKGLEWVA
	HFR3 (protein)	75	RFTISRDN AKNTLYLQMNSLRAEDTALYYCAR
	HFR4 (protein)	76	WGQGLTVTVSS
Girentuximab	HFR1 (DNA)	77	GAGGTGCAGCTGGTGGAGTCTGGGGGAGGCTTGGT CAAGCCTGGAGGATCCCTGAGACTCTCCTGTGCAG CCTCTGGATTACCTTCAGT
	HFR2 (DNA)	78	TGGGTGCGCCAGGCTCCAGGGAAGGGGCTGGAGT GGGTTGCC
	HFR3 (DNA)	79	CGATTACCATCTCCAGGGACAACGCCAAGAACACC CTGTATCTGCAATGAACAGCCTGAGGGCCGAGGA CACGGCCCTGTATTACTGTGCGAGA
	HFR4 (DNA)	80	TGGGGCCAGGGCACCCCTGGTCACTGTCTCCTCA
	LCDR1 (protein)	81	KASQNVVSAVA
	LCDR2 (protein)	82	SASNRYT
Chimeric Variable light chain	LCDR3 (protein)	83	QQYSNYPWT

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	VL (protein)	84	DIVMTQSQRFMSTTVGDRVSITCKASQNVVSAVAWYQ QKPGQSPKLLIYSASNRYTGVPDRFTGSGSGTDFTLTI SNMQSEDLADFFCQQYSNYPWTFGGGTKLEIK
	LCDR1 (DNA)	85	AAGGCCAGTCAGAATGTGGTTTCTGCTGTTGCC
	LCDR2 (DNA)	86	TCAGCATCCAATCGGTACACT
	LCDR3 (DNA)	87	CAACAATATAGCAACTATCCGTGGACG
	VL (DNA)	88	GACATTGTGATGACCCAGTCTCAAAGATTCATGTCC ACAACAGTAGGAGACAGGGTCAGCATCACCTGCAA GGCCAGTCAGAATGTGGTTTCTGCTGTTGCCTGGTA TCAACAGAAACCAGGACAATCTCCTAACTACTGATT TACTCAGCATCCAATCGGTACACTGGAGTCCCTGAT CGCTTCACAGGCAGTGGATCTGGGACAGATTTCACT CTCACCATTAGCAATATGCAGTCTGAAGACCTGGCT GATTTTTTCTGTCAACAATATAGCAACTATCCGTGGA CGTTCGGTGGAGGCACCAAGCTGGAAATCAAA
	LFR1 (protein)	89	DIVMTQSQRFMSTTVGDRVSITC
	LFR2 (protein)	90	WYQQKPGQSPKLLIY
	LFR3 (protein)	91	GVPDRFTGSGSGTDFTLTISNMQSEDLADFFC
	LFR4 (protein)	92	FGGGTKLEIK
	LFR1 (DNA)	93	GACATTGTGATGACCCAGTCTCAAAGATTCATGTCC ACAACAGTAGGAGACAGGGTCAGCATCACCTGC
	LFR2 (DNA)	94	TGGTATCAACAGAAACCAGGACAATCTCCTAACTA CTGATTTAC
	LFR3 (DNA)	95	GGAGTCCCTGATCGCTTCACAGGCAGTGGATCTGG GACAGATTTCACTCTCACCATTAGCAATATGCAGTCT GAAGACCTGGCTGATTTTTTCTGT
	LFR4 (DNA)	96	TTCGGTGGAGGCACCAAGCTGGAAATCAAA
Girentuximab Humanised Vk1 Variable light chain	LCDR1 (protein)	97	KASQNVVSAVA
	LCDR2 (protein)	98	SASNRYT
	LCDR3 (protein)	99	QQYSNYPWT
	VL (protein)	100	DIVMTQSPSFLSASVGDRVITITCKASQNVVSAVAWYQ QKPGQAPKLLIYSASNRYTGVPDRFTGSGSGTDFTLTI SSLQSEDLADYFCQQYSNYPWTFGGGTKVEIK
	LCDR1 (DNA)	101	AAGGCAAGTCAGAACGTGGTGAGTGCTGTGGCC

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	LCDR2 (DNA)	102	AGCGCCTCCAACAGGTACACC
	LCDR3 (DNA)	103	CAACAGTACAGCAATTACCCTTGGACG
	VL (DNA)	104	GACATCGTGATGACCCAGTCTCCATCCTTCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGCAAG GCAAGTCAGAACGTGGTGAGTGCTGTGGCCTGGTA TCAGCAGAAACCAGGGCAGGCTCCTAAGCTCCTGAT CTATAGCGCCTCCAACAGGTACACCGGGTCCCAG ACAGGTTACACGGCAGTGATCTGGGACAGATTTCA CTCTACCATCAGCAGCCTGCAGAGCGAAGATCTG GCAGACTATTTCTGTCAACAGTACAGCAATTACCCTT GGACGTTTCGGCGGCGGGACCAAGGTGGAAATCAAA
	LFR1 (protein)	105	DIVMTQSPSFLSASVGDRTITC
	LFR2 (protein)	106	WYQQKPGQAPKLLIY
	LFR3 (protein)	107	GVPDRFTGSGSGTDFTLTISSLQSEDLADYFC
	LFR4 (protein)	108	FGGGTKVEIK
	LFR1 (DNA)	109	GACATCGTGATGACCCAGTCTCCATCCTTCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGC
	LFR2 (DNA)	110	TGGTATCAGCAGAAACCAGGGCAGGCTCCTAAGCT CCTGATCTAT
	LFR3 (DNA)	111	GGGGTCCCAGACAGGTTACCGGCAGTGATCTGG GACAGATTTCACTCTCACCATCAGCAGCCTGCAGAG CGAAGATCTGGCAGACTATTTCTGT
	LFR4 (DNA)	112	TTCGGCGGCGGGACCAAGGTGGAAATCAAA
	LFR4 (DNA)	112	TTCGGCGGCGGGACCAAGGTGGAAATCAAA
Girentuximab Humanised Vk2 Variable light chain	LCDR1 (protein)	113	KASQNVVSAVA
	LCDR2 (protein)	114	SASNRYT
	LCDR3 (protein)	115	QQYSNYPWT
	VL (protein)	116	DIVMTQSPSSLSASVGDRTITCKASQNVVSAVAWYQ QKPGQAPRLLIYSASNRYTGVPDRFTGSGSGTDFTLTI SSLQAEDLADYFCQQYSNYPWTFGGGKVEIK
	LCDR1 (DNA)	117	AAGGCAAGTCAGAACGTGGTGAGTGCTGTGGCC
	LCDR2 (DNA)	118	AGCGCCTCCAACAGGTACACC
	LCDR3 (DNA)	119	CAACAGTACAGCAATTACCCTTGGACG

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	VL (DNA)	120	GACATCGTGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGCAAG GCAAGTCAGAACGTGGTGAGTGCTGTGGCCTGGTA TCAGCAGAAACCAGGGCAGGCTCCTAGGCTCCTGA TCTATAGCGCCTCCAACAGGTACACCGGGGTCCCA GACAGGTTACCGGCAGTGGATCTGGGACAGATTT CACTCTCACCATCAGCAGCCTGCAGGCCGAAGATCT GGCAGACTATTTCTGTCAACAGTACAGCAATTACCC TTGGACGTTTCGGCGGCGGGACCAAGGTGGAAATCA AA
	LFR1 (protein)	121	DIVMTQSPSSLSASVGDRVITIC
	LFR2 (protein)	122	WYQQKPGQAPRLIY
	LFR3 (protein)	123	GVPDRFTGSGSGTDFTLTISSLQAEDLADYFC
	LFR4 (protein)	124	FGGGTKVEIK
	LFR1 (DNA)	125	GACATCGTGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGC
	LFR2 (DNA)	126	TGGTATCAGCAGAAACCAGGGCAGGCTCCTAGGCT CCTGATCTAT
	LFR3 (DNA)	127	GGGGTCCCAGACAGGTTACCGGCAGTGGATCTGG GACAGATTTCACTCTCACCATCAGCAGCCTGCAGGC CGAAGATCTGGCAGACTATTTCTGT
	LFR4 (DNA)	128	TCGGCGGCGGGACCAAGGTGGAAATCAAA
Girentuximab Humanised Vk3 Variable light chain	LCDR1 (protein)	129	KASQNVVSAVA
	LCDR2 (protein)	130	SASNRYT
	LCDR3 (protein)	131	QQYSNYPWT
	VL (protein)	132	DIQMTQSPSSLSASVGDRVITICKASQNVVSAVAWYQ QKPGQAPRLIYSASNRYTGVPDRFSGSGTDFTLTI SSLQAEDLADYFCQQYSNYPWTFGGGTKVEIK
	LCDR1 (DNA)	133	AAGGCAAGTCAGAACGTGGTGAGTGCTGTGGCC
	LCDR2 (DNA)	134	AGCGCCTCCAACAGGTACACC
	LCDR3 (DNA)	135	CAACAGTACAGCAATTACCCTTGGACG

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	VL (DNA)	136	GACATCCAGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGCAAG GCAAGTCAGAACGTGGTGAGTGCTGTGGCCTGGTA TCAGCAGAAACCAGGGCAGGCTCCTAGGCTCCTGA TCTATAGCGCCTCCAACAGGTACACCGGGGTCCCA GACAGGTTTCAGCGGCAGTGGATCTGGGACAGATTT CACTCTCACCATCAGCAGCCTGCAGGCCGAAGATCT GGCAGACTATTTCTGTCAACAGTACAGCAATTACCC TTGGACGTTTCGGCGGCGGGACCAAGGTGGAAATCA AA
	LFR1 (protein)	137	DIQMTQSPSSLSASVGDRVITIC
	LFR2 (protein)	138	WYQQKPGQAPRLIY
	LFR3 (protein)	139	GVPDRFSGSGSGTDFTLTISSLQAEDLADYFC
	LFR4 (protein)	140	FGGGTKVEIK
	LFR1 (DNA)	141	GACATCCAGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGC
	LFR2 (DNA)	142	TGGTATCAGCAGAAACCAGGGCAGGCTCCTAGGCT CCTGATCTAT
	LFR3 (DNA)	143	GGGGTCCCAGACAGGTTTCAGCGGCAGTGGATCTGG GACAGATTTCACTCTCACCATCAGCAGCCTGCAGGC CGAAGATCTGGCAGACTATTTCTGT
	LFR4 (DNA)	144	TTCGGCGGCGGGACCAAGGTGGAAATCAAA
Girentuximab Humanised Vk4 Variable light chain	LCDR1 (protein)	145	KASQNVVSAVA
	LCDR2 (protein)	146	SASNRYT
	LCDR3 (protein)	147	QQYSNYPWT
	VL (protein)	148	DIQMTQSPSSLSASVGDRVITICKASQNVVSAVAWYQ QKPGQAPRLIYSASNRYTGVPDRFSGSGSGTDFTLTI SSLQAEDLADYYCQQYSNYPWTFGGGTKVEIK
	LCDR1 (DNA)	149	AAGGCAAGTCAGAACGTGGTGAGTGCTGTGGCC
	LCDR2 (DNA)	150	AGCGCCTCCAACAGGTACACC
	LCDR3 (DNA)	151	CAACAGTACAGCAATTACCCTTGGACG

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	VL (DNA)	152	GACATCCAGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGCAAG GCAAGTCAGAACGTGGTGAGTGCTGTGGCCTGGTA TCAGCAGAAACCAGGGCAGGCTCCTAGGCTCCTGA TCTATAGCGCCTCCAACAGGTACACCGGGGTCCCA GACAGGTTTCAGCGGCAGTGGATCTGGGACAGATTT CACTCTCACCATCAGCAGCCTGCAGGCCGAAGATCT GGCAGACTATTACTGTCAACAGTACAGCAATTACCC TTGGACGTTTCGGCGGCGGGACCAAGGTGGAAATCA AA
	LFR1 (protein)	153	DIQMTQSPSSLSASVGDRVITIC
	LFR2 (protein)	154	WYQQKPGQAPRLIY
	LFR3 (protein)	155	GVPDRFSGSGSGTDFTLTISSLQAEDLADYYC
	LFR4 (protein)	156	FGGGTKVEIK
	LFR1 (DNA)	157	GACATCCAGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGC
	LFR2 (DNA)	158	TGGTATCAGCAGAAACCAGGGCAGGCTCCTAGGCT CCTGATCTAT
	LFR3 (DNA)	159	GGGGTCCCAGACAGGTTTCAGCGGCAGTGGATCTGG GACAGATTTCACTCTCACCATCAGCAGCCTGCAGGC CGAAGATCTGGCAGACTATTACTGT
	LFR4 (DNA)	160	TCGGCGGCGGGACCAAGGTGGAAATCAAA
Girentuximab Humanised Vk5 Variable light chain	LCDR1 (protein)	161	KASQNVVSAVA
	LCDR2 (protein)	162	SASNRYT
	LCDR3 (protein)	163	QQYSNYPWT
	VL (protein)	164	DIQMTQSPSSLSASVGDRVITICKASQNVVSAVAWYQ QKPGQAPRRLIYSASNRYTGVPDRFSGSGSGTDFTLTI SSLQAEDLADYYCQQYSNYPWTFGGGTKVEIK
	LCDR1 (DNA)	165	AAGGCAAGTCAGAACGTGGTGAGTGCTGTGGCC
	LCDR2 (DNA)	166	AGCGCCTCCAACAGGTACACC
	LCDR3 (DNA)	167	CAACAGTACAGCAATTACCCTTGGACG

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
	VL (DNA)	168	GACATCCAGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGCAAG GCAAGTCAGAACGTGGTGAGTGCTGTGGCCTGGTA TCAGCAGAAACCAGGGCAGGCTCCTAGGAGGCTGA TCTATAGCGCCTCCAACAGGTACACCGGGGTCCCA GACAGGTTTCAGCGGCAGTGGATCTGGGACAGATTT CACTCTCACCATCAGCAGCCTGCAGGCCGAAGATCT GGCAGACTATTACTGTCAACAGTACAGCAATTACCC TTGGACGTTTCGGCGGCGGGACCAAGGTGGAAATCA AA
	LFR1 (protein)	169	DIQMTQSPSSLSASVGDRTITC
	LFR2 (protein)	170	WYQQKPGQAPRRLIY
	LFR3 (protein)	171	GVPDRFSGSGSGTDFTLTISSLQAEDLADYYC
	LFR4 (protein)	172	FGGGTKVEIK
	LFR1 (DNA)	173	GACATCCAGATGACCCAGTCTCCATCCAGCCTGTCT GCATCTGTAGGAGACAGAGTCACCATCACTTGC
	LFR2 (DNA)	174	TGGTATCAGCAGAAACCAGGGCAGGCTCCTAGGAG GCTGATCTAT
	LFR3 (DNA)	175	GGGGTCCCAGACAGGTTTCAGCGGCAGTGGATCTGG GACAGATTTCACTCTCACCATCAGCAGCCTGCAGGC CGAAGATCTGGCAGACTATTACTGT
	LFR4 (DNA)	176	TTCGGCGGCGGGACCAAGGTGGAAATCAAA
Girentuximab unmodified human IgG1 constant chain region	IgG1 HC (protein)	177	ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLSSVTVPSST LGTQTYICNVNHKPSNTKVDKKVEPKSCDKTHTCPPC PAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVDVS HEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVV SVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAV EWESNGQPENNYKTPPVLDSDGSFFLYSKLTVDKSR WQQGNVFSCSVMHEALHNHYTQKSLSLSPGK

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
Girentuximab modified human IgG1 constant chain region	IgG1 H310A H435Q HC (protein)	178	ASTKGPSVFPLAPSSKSTSGGTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLSSVTVPS LGTQTYICNVNHKPSNTKVDKKVEPKSCDKTHTCPPC PAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVDV HEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVV SVLTVLAQDWLNGKEYKCKVSNKALPAPIEKTISKAKG QPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAV EWESNGQPENNYKTTTPVLDSDGSFFLYSKLTVDKSR WQQGNVFSCSVMHEALHNQYTKQSLSLSPGK
Girentuximab modified human IgG4 constant chain region	IgG4 S228P L235E HC (protein)	179	ASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLSSVTVPS LGTKTYTCNVDHKPSNTKVDKRVESKYGPPCPPCAP EFEGGPSVFLFPPKPKDTLMISRTPEVTCVVDV PEVQFNWYVDGVEVHNAKTKPREEQFNSTYRVVSVLT VLHQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPR EPQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIAVEW ESNGQPENNYKTTTPVLDSDGSFFLYSRLTVDKSRWQ EGNVFSCSVMHEALHNHYTQKSLSLGLGK
Girentuximab modified human IgG4 constant chain region	IgG4 S228P L235E H310A H435Q HC (protein)	180	ASTKGPSVFPLAPCSRSTSESTAALGCLVKDYFPEPVT VSWNSGALTSGVHTFPAVLQSSGLYSLSSVTVPS LGTKTYTCNVDHKPSNTKVDKRVESKYGPPCPPCAP EFEGGPSVFLFPPKPKDTLMISRTPEVTCVVDV PEVQFNWYVDGVEVHNAKTKPREEQFNSTYRVVSVLT VLAQDWLNGKEYKCKVSNKGLPSSIEKTISKAKGQPRE PQVYTLPPSQEEMTKNQVSLTCLVKGFYPSDIAVEW SNGQPENNYKTTTPVLDSDGSFFLYSRLTVDKSRWQE GNVFSCSVMHEALHNQYTKQSLSLGLGK
Girentuximab kappa light chain constant region	Human κ LC constant region	181	RTVAAPSVFIFPPSDEQLKSGTASVVCLLNNFYPREAK VQWKVDNALQSGNSQESVTEQDSKDSSTYSSTLTLS KADYEEKHKVYACEVTHQGLSSPVTKSFNRGEC

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
Girentuximab native IgG1 allotype Gm (1,17) VH4 heavy chain	IgG1 VH4 heavy chain	182	EVQLVESGGGLVKPGGSLRLSCAASGFTFSNYYMSW VRQAPGKGLELVAAINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMNSLRAEDTALYYCARHRSRGYFSMDYWG QGTLTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLV KDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTVPSSSLGTQTYICNVNHKPSNTKVDKKVEPKSCD KTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEV TCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQ YNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIE KTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKG FYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYS KLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLS PGK
hGirentuximab IgG1 allotype Gm (1,17) RADmAb VH4 heavy chain	FcRn-null IgG1 allotype H130A H435Q	183	EVQLVESGGGLVKPGGSLRLSCAASGFTFSNYYMSW VRQAPGKGLELVAAINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMNSLRAEDTALYYCARHRSRGYFSMDYWG QGTLTVSSASTKGPSVFPLAPSSKSTSGGTAALGCLV KDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTVPSSSLGTQTYICNVNHKPSNTKVDKKVEPKSCD KTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEV TCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQ YNSTYRVVSVLTVLAQDWLNGKEYKCKVSNKALPAPIE KTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKG FYPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYS KLTVDKSRWQQGNVFSCSVMHEALHNQYTQKSLSLS PGK
hGirentuximab IgG4 RADmAb VH4 heavy chain	FcR γ -null IgG4 S228P L235E	184	EVQLVESGGGLVKPGGSLRLSCAASGFTFSNYYMSW VRQAPGKGLELVAAINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMNSLRAEDTALYYCARHRSRGYFSMDYWG QGTLTVSSASTKGPSVFPLAPCSRSTSESTAALGCLV KDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTVPSSSLGTQTYTCNVDHKPSNTKVDKRVESKYG PPCPPCPAPEFEGGPSVFLFPPKPKDTLMISRTPEVTC VVVDVSDQEDPEVQFNWYVDGVEVHNAKTKPREEQFN STYRVVSVLTVLHQDWLNGKEYKCKVSNKGLPSSIEKT ISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSR LTVDKSRWQEGNVFSCSVMHEALHNHYTQKSLSLSLG K

Antibody ID	Region	SEQ ID NO:	Amino acid or nucleotide sequence
hGirentuximab IgG4 RADmAb VH4 heavy chain	FcRn+FcRγ-null IgG4 S228P L235E H310A H435Q	185	EVQLVESGGGLVKPGGSLRLSCAASGFTFSNYYMSW VRQAPGKGLLELVAAINSDGGITYYLDTVKGRFTISRDN AKNTLYLQMNSLRAEDTALYYCARHRSGYFSMDYWG QGTLVTVSSASTKGPSVFPLAPCSRSTSESTAALGCLV KDYFPEPVTVSWNSGALTSGVHTFPAVLQSSGLYSLS SVVTVPSSSLGTKTYTCNVDHKPSNTKVDKRVESKYG PPCPPCPAPEFEGGPSVFLFPPKPKDTLMISRTPEVTC VVVDVSQEDPEVQFNWYVDGVEVHNAKTKPREEQFN STYRVVSVLTVLAQDWLNGKEYKCKVSNKGLPSSIEKT ISKAKGQPREPQVYTLPPSQEEMTKNQVSLTCLVKGF YPSDIAVEWESNGQPENNYKTPPVLDSDGSFFLYSR LTVDKSRWQEGNVFSCSVMHEALHNQYTKQKSLSLSL GK
hGirentuximab Vκ Light Chain	Vκ Light Chain	186	DIQMTQSPSSLSASVGDRVTITCKASQNVVSAVAWYQ QKPGQAPRLLIYSASNRYTGVPDRFSGSGSGTDFTLTI SSLQAEDLADYYCQQYSNPWTFGGGTKVEIKRTVAA PSVFIFPPSDEQLKSGTASVVCLLNFPYPREAKVQWKV DNALQSGNSQESVTEQDSKDYSLSTLTLSKADYE KHKVYACEVTHQGLSSPVTKSFNRGEC

[0139] In further embodiments, the agent for binding to CAIX may comprise BCA-356, a bispecific antibody comprising affinity matured humanised anti-CAIX antibody linked to attenuated subunits of IL-12 fused to each of the heavy chains of the anti-CAIX antibody at the C-terminus via a linker into a “knobs in hole” format (as described in Nair et al., Journal for ImmunoTherapy of Cancer, 10:S2).

[0140] In an especially preferred embodiment, the agent for binding to CAIX is ¹⁷⁷Lu-Girentuximab. ¹⁷⁷Lu-Girentuximab is a chimeric monoclonal antibody (INN name: girentuximab (GTX), also known as cG250 and TLX250) with specificity for CAIX antigen, radiolabelled with the lutetium 177 via a DOTA linker.

Constant regions

[0141] In preferred embodiments, an antibody and/or antigen binding fragment thereof as described herein for use in the present invention may comprise a constant region of an antibody. This includes antigen binding fragments of an antibody fused to an Fc.

[0142] Sequences of constant regions useful for producing the antibodies or antigen binding fragment thereof as described herein may be obtained from a number of different sources. In some examples, the constant region or portion thereof of the protein is derived from a human antibody. The constant region or portion thereof may be derived from any antibody class, including IgM, IgG, IgD, IgA and IgE, and any antibody isotype, including IgG1, IgG2, IgG3 and IgG4. In one example, the constant region is human isotype IgG4 or a stabilized IgG4 constant region.

[0143] The neonatal Fc-receptor (FcRn) is important for the metabolic fate of antibodies of the IgG class *in vivo*. The FcRn functions to salvage IgG from the lysosomal degradation pathway, resulting in reduced clearance and increased half-life. It is a heterodimeric protein consisting of two polypeptides: a 50 kDa class I major histocompatibility complex-like protein (α -FcRn) and a 15 kDa β 2-microglobulin (β 2m). FcRn binds with high affinity to the CH2-CH3 portion of the Fc-region of an antibody of the class IgG. The interaction between an antibody of the class IgG and the FcRn is pH dependent and occurs in a 1 :2 stoichiometry, i.e. one IgG antibody molecule can interact with two FcRn molecules via its two heavy chain Fc-region polypeptides (see e.g. Huber, A.H., et al, J. Mol. Biol. 230 (1993) 1077-1083).

[0144] Thus, an IgG's *in vitro* FcRn binding properties/characteristics are indicative of its *in vivo* pharmacokinetic properties in the blood circulation. In the interaction between the FcRn and the Fc-region of an antibody of the IgG class different amino acid residues of the heavy chain CH2- and CH3 -domain are participating.

[0145] Different mutations that influence the FcRn binding and therewith the half-live in the blood circulation are known. Fc-region residues critical to the mouse Fc-region-mouse FcRn interaction have been identified by site-directed mutagenesis (see e.g. Dall'Acqua, W.F., et al. J. Immunol 169 (2002) 5171-5180). Residues Ile253, His310, His433, Asn434 and His435 (numbering according to EU index numbering system) are involved in the interaction (Medesan, C, et al., Eur. J. Immunol. 26 (1996) 2533-2536; Firan, M., et al, Int. Immunol. 13 (2001) 993-1002; Kim, J.K., et al, Eur. J. Immunol. 24 (1994) 542-548). (Using the Kabat system, the relevant residues are Ile266, His329, His464, Asn465 and His466). Residues Ile253, His310, and His435 were found to be critical for the interaction of human Fc-region with murine FcRn (Kim, J.K., et al, Eur. J. Immunol. 29 (1999) 2819-2885).

[0146] More specifically, the antibody or antigen binding protein may comprise one or more amino acid substitutions that decrease the half-life of the protein. For example, the antibody or antigen binding fragment thereof may comprise a Fc region comprising one or more amino acid substitutions that decrease the affinity of the Fc region for the neonatal Fc region (FcRn).

Preferred Modifications

[0147] In any embodiment, the antibody or antigen binding fragment thereof (for example a G250 antibody or variant thereof as described herein) is a modified IgG antibody or fragment thereof, comprising a heavy chain constant region having one or more amino acid substitutions compared to a wild-type antibody of the class IgG, wherein the one or more amino acid substitutions reduce the affinity of the antibody for the neonatal Fc receptor (FcRn), thereby reducing the serum half-life of the modified antibody compared to a wild-type antibody of class IgG.

[0148] In one embodiment, the one or more amino acid substitutions are selected from substitutions in the heavy chain constant region 2 (CH2) of the IgG molecule, reducing the affinity of the IgG molecule for FcRn. Alternatively, the one or more amino acid substitutions may be in the heavy chain constant region 3 (CH3) of the IgG molecule, thereby reducing the affinity of the IgG molecule for FcRn. Still further, the amino acid substitutions may include at least one substitution in the CH2 region, and at least one substitution in the CH3 region of the IgG molecule, whereby the substitutions reduce the affinity of the IgG for FcRn.

[0149] In certain preferred embodiments, the one or more amino acid substitutions may be at one or more of residues His310, His433, His435, His436, or Ile253 of IgG. Preferably, the amino acid substitutions comprise a substitution in the heavy chain constant region at positions His310 or at His435. More preferably, the amino acid substitutions that reduce the affinity of the antibody for FcRn are at both His310 and His435.

[0150] In other preferred embodiments, the antibody and/or antigen binding fragment thereof has a constant region substantially identical to a naturally occurring class IgG antibody constant region wherein at least one amino acid residue selected from the group consisting of residues His310, His435, and Ile253 is different from that present in the

naturally occurring class IgG antibody, thereby altering FcRn binding affinity and/or serum half-life of said antibody relative to the naturally occurring antibody. In preferred embodiments, the naturally occurring class IgG antibody comprises a heavy chain constant region of a human IgG1, IgG2, IgG2M3, IgG3 or IgG4 molecule.

[0151] Also in preferred embodiments, amino acid residue 310 and/or residue 435 from the heavy chain constant region of the antibody having a constant region substantially identical to the naturally occurring class IgG antibody is any amino acid that is not histidine and which reduces the affinity of the constant region for FcRn. For example, the amino acid at residue 310 and/or 435 may be alanine, glutamic acid, aspartic acid, leucine, isoleucine, arginine, proline, glutamine, methionine, serine, threonine, lysine, asparagine, phenylalanine, tyrosine, tryptophan, cysteine, valine or glycine.

[0152] The amino acid substitutions may include substitution from a histidine residue to: alanine, glutamine, glutamic acid or aspartic acid. Preferably, the amino acid substitution at His310 is to alanine. Preferably the amino acid substitution at His435 is to glutamine. Preferably, the amino acid substitution at Ile253 is alanine.

[0153] In a preferred embodiment of the present invention, the binding affinity for FcRn and/or the serum half-life of the modified antibody is decreased by at least about 30%, 50%, 80%, 2-fold, 3-fold, 4-fold, 5-fold, 10-fold, 15-fold, 20-fold, 25-fold, 30-fold, 40-fold, 50-fold, 60-fold, 70-fold, 80-fold, 90-fold, or 100-fold. In a preferred embodiment of the present invention, the binding affinity for FcRn and/or the serum half-life of said modified antibody is reduced by at least about 20%, 30%, 40%, 50%, 60%, 70%, 80%, 85%, 90%, 95%, 97%, 98%, or 99%.

[0154] In addition, the antibodies or fragments thereof for use according to the present invention may be modified to comprise one or more mutations which modify the affinity of the antibodies for any one or more Fc gamma receptors. For example, the one or more amino acid modifications change the affinity of the antibody constant domain, Fc region, or Fc gamma receptor binding fragment, for any one or more Fc gamma receptors.

[0155] In certain embodiments, the modified antibody or antigen binding fragment thereof retains the ability to bind to one or more Fc-gamma receptors and accordingly, in certain embodiments the modified antibody retains the ability to stimulate effector responses (including ADCC). In one example, the Fc region of the constant region

contains one or more amino acid substitutions that modulate effector function, including increasing effector function compared to a wild-type IgG.

[0156] In one example, the Fc region of the constant region has a reduced ability to induce effector function, e.g., compared to a native or wild-type human IgG1 or IgG3 Fc region. In one example, the effector function is antibody-dependent cell-mediated cytotoxicity (ADCC) and/or antibody-dependent cell-mediated phagocytosis (ADCP) and/or complement-dependent cytotoxicity (CDC). Methods for assessing the level of effector function of an Fc region containing protein are known in the art and/or described herein.

[0157] In one example, the amino acid substitution that modifies that ability of the antibody to induce effector function is an amino acid substitution at residue Ile253 from the heavy chain constant region. In one example, the substitution is to any amino acid selected from be alanine, glutamic acid, aspartic acid, leucine, isoleucine, arginine, proline, glutamine, methionine, serine, threonine, lysine, asparagine, phenylalanine, tyrosine, tryptophan, cysteine, valine or glycine, wherein the substitution reduces the ability of the antibody to induce effector function. In preferred embodiments, the substitution from Ile at residue 253 is to arginine, proline, glutamic acid or aspartate, more preferably alanine.

[0158] In one example, the Fc region is an IgG4 Fc region (i.e., from an IgG4 constant region), e.g., a human IgG4 Fc region. Sequences of suitable IgG4 Fc regions will be apparent to the skilled person and/or available in publicly available databases (e.g., available from National Center for Biotechnology Information).

[0159] In one example, the constant region is a stabilized IgG4 constant region. The term "stabilized IgG4 constant region" will be understood to mean an IgG4 constant region that has been modified to reduce Fab arm exchange or the propensity to undergo Fab arm exchange or formation of a half-antibody or a propensity to form a half antibody. "Fab arm exchange" refers to a type of protein modification for human IgG4, in which an IgG4 heavy chain and attached light chain (half-molecule) is swapped for a heavy-light chain pair from another IgG4 molecule. Thus, IgG4 molecules may acquire two distinct Fab arms recognizing two distinct antigens (resulting in bispecific molecules). Fab arm exchange occurs naturally in vivo and can be induced in vitro by purified blood cells or reducing agents such as reduced glutathione. A "half antibody" forms when an IgG4

antibody dissociates to form two molecules each containing a single heavy chain and a single light chain.

[0160] In one example, a stabilized IgG4 constant region comprises a proline at position 241 of the hinge region according to the system of Kabat (Kabat et al., Sequences of Proteins of Immunological Interest Washington DC United States Department of Health and Human Services, 1987 and/or 1991). This position corresponds to position 228 of the hinge region according to the EU numbering system. In human IgG4, this residue is generally a serine. Following substitution of the serine for proline, the IgG4 hinge region comprises a sequence CPPC. In this regard, the skilled person will be aware that the "hinge region" is a proline-rich portion of an antibody heavy chain constant region that links the Fc and Fab regions that confers mobility on the two Fab arms of an antibody. The hinge region includes cysteine residues which are involved in inter-heavy chain disulfide bonds. It is generally defined as stretching from Glu226 to Pro243 of human IgG1 according to the numbering system of Kabat (or Glu216 to Pro230 using the EU index). Hinge regions of other IgG isotypes may be aligned with the IgG1 sequence by placing the first and last cysteine residues forming inter-heavy chain disulphide (S-S) bonds in the same positions (see for example WO2010/080538).

[0161] In alternative embodiments, the one or more amino acid modifications which reduce the affinity for the FcRn receptor also reduce the affinity for the Fc gamma receptors. The modified antibody or antigen binding fragment thereof may further comprise one or more amino acid substitutions compared a wild-type antibody of the class IgG, wherein the amino acid substitutions further reduce the affinity of the antibody for one or more Fc gamma receptors.

[0162] In a further embodiment, the modified antibody or antigen binding fragment thereof further comprises one or more amino acid substitutions compared a wild-type antibody of the class IgG, wherein the amino acid substitutions increase the stability of the CH1-CH2 hinge region in the modified antibody compared to a wild-type antibody of the class IgG.

[0163] In any embodiment, the heavy chain constant region of the antibody or antigen binding protein comprises amino acid substitutions at both His310 and His435. The antibody may also comprise amino acid substitutions at residues equivalent to Ser228 and Leu235 of the constant heavy chain region.

[0164] In any embodiment, the antibody or antigen binding fragment thereof comprises mutations at Ser228, Leu235, His310 and His435. Preferably, the amino acid modifications are Ser228Pro, Leu235Glu, His310Ala and His435Gln.

[0165] Additional examples of stabilized IgG4 antibodies are antibodies in which arginine at position 409 in a heavy chain constant region of human IgG4 (according to the EU numbering system) is substituted with lysine, threonine, methionine, or leucine (e.g., as described in WO2006/033386). The Fc region of the constant region may additionally or alternatively comprise a residue selected from the group consisting of: alanine, valine, glycine, isoleucine and leucine at the position corresponding to 405 (according to the EU numbering system). Optionally, the hinge region comprises a proline at position 241 (i.e., a CPPC sequence) (as described above).

[0166] In another example, the Fc region is a region modified to have reduced effector function, i.e., a “non-immunostimulatory Fc region”. For example, the Fc region is an IgG1 Fc region comprising a substitution at one or more positions selected from the group consisting of 268, 309, 330 and 331. In another example, the Fc region is an IgG1 Fc region comprising one or more of the following changes E233P, L234V, L235A and deletion of G236 and/or one or more of the following changes A327G, A330S and P331S (Armour et al., Eur J Immunol. 29:2613-2624, 1999; Shields et al., J Biol Chem. 276(9):6591-604, 2001). Additional examples of non-immunostimulatory Fc regions are described, for example, in Dall'Acqua et al., J Immunol. 177 : 1129-1138 2006; and/or Hezareh J Virol ;75: 12161-12168, 2001).

[0167] In another example, the Fc region is a chimeric Fc region, e.g., comprising at least one CH2 domain from an IgG4 antibody and at least one CH3 domain from an IgG1 antibody, wherein the Fc region comprises a substitution at one or more amino acid positions selected from the group consisting of 240, 262, 264, 266, 297, 299, 307, 309, 323, 399, 409 and 427 (EU numbering) (e.g., as described in WO2010/085682). Exemplary substitutions include 240F, 262L, 264T, 266F, 297Q, 299A, 299K, 307P, 309K, 309M, 309P, 323F, 399S, and 427F.

[0168] Preferably, the antibody or antigen binding fragment thereof comprises a heavy chain constant region comprising the sequence as set forth in any one of SEQ ID NOs: 177 to 180, preferably as set forth in SEQ ID NO: 178.

[0169] In a still further embodiment, the antibody or antigen binding fragment thereof preferably comprises a heavy chain comprising the sequence set forth in any one of SEQ ID NOs: 182 to 185, preferably as set forth in SEQ ID NO: 183.

[0170] In any embodiment, the antibody or antigen binding fragment thereof comprises a light chain constant region comprising the amino acid sequence as set forth in SEQ ID NO: 181. Preferably, the antibody or antigen binding protein comprises a light chain comprising the amino acid sequence as set forth in SEQ ID NO: 186.

[0171] In any embodiment, the antibody or antigen binding fragment thereof comprises the sequence set forth in SEQ ID NO: 183 and the sequence set forth in SEQ ID NO: 186.

[0172] In one embodiment, the antibody or antigen binding fragment thereof comprises: a VH comprising a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 36 or 52 and the VL comprises a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 116, 132 or 148.

[0173] Preferably, the VH comprises a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 36 or 52 and a VL comprising a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 132 or 148.

[0174] More preferably, the VH comprises a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 36 and the VL comprises a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 148.

[0175] Alternatively, the VH comprises a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 52 and the VL comprises a sequence at least about 95% or 96% or 97% or 98% or 99% identical to, or comprises a sequence set forth in SEQ ID NO: 132 or 148, preferably the sequence set forth in SEQ ID NO: 148.

Radiolabelled antibodies

[0176] The skilled person will be familiar with standard methods for conjugating a detectable moiety such as a radionuclide (radiolabel) to an antibody or antigen binding fragment thereof.

[0177] As used herein, the term radionuclide may be used interchangeably with the term radioisotope and radiolabel.

[0178] In any embodiment of the invention, radiolabel may be conjugated to the agent (such as girentuximab or antigen binding fragment thereof), directly via binding to single or multiple amino acid residues in the protein (e.g. halogenation of tyrosine residues) or indirectly (via a chelating agent or prosthetic group or linker) linked to the radioisotope (ie the radiolabel).

[0179] Examples of suitable chelating agents or linkers may be selected from the group consisting of: TMT (6,6"-bis[N,N",N""-tetra(carboxymethyl)aminomethyl]-4'-(3-amino-4-methoxyphenyl)-2,2':6',2"-terpyridine), DOTA (1, 4,7,10-tetraazacyclododecane-NN',N"(N""-tetraacetic acid, also known as tetraxetan), TCMC (the tetra-primary amide of DOTA), DO3A (1,4,7,10-Tetraazacyclododecane-1,4,7-tris(acetic acid)-10-(2-thioethyl)acetamide), CB-DO2A (4,10-bis(carboxymethyl)-1,4,7,10-tetraazabicyclo[5.5.2]tetradecan), NOTA (1,4,7-triazacyclononane-triacetic acid) Diamsar (3,6,10,13,16,19-hexaazabicyclo[6.6.6]eicosane-1,8-diamine), DTPA (Pentetic acid or diethylenetriaminepentaacetic acid), CHX-A"-DTPA ([[(R)-2-Amino-3-(4-isothiocyanatophenyl)propyl]-trans-(S,S)-cyclohexane-1,2-diamine-pentaacetic acid), TETA (1,4,8,11-tetraazacyclotetradecane-1,4,8), 11-tetraacetic acid, Te2A (4,11 - bis(carboxymethyl)-1,4,8,11-tetraazabicyclo[6.6.2]hexadecane), HBED, DFO (Desferrioxamine), DFOsq (DFO-squaramide) and HOPO (3,4,3-(LI-1,2-HOPO) or other chelating agent as described herein. Other known chelating moieties include 3p-C-NETA ({4-[2-(bis-carboxy-methylamino)-5-(4-nitrophenyl)pentyl]-7-carbo-xymethyl-[1,4,7]triazanonan-1-yl} acetic acid), 5p-C-NETA (2-({1-[4,7-bis(carboxymethyl)-1,4,7-triazanonan-1-yl]-7-(4-nitrophenyl)heptan-2-yl}(carbo-xymethyl) amino)acetic acid), NOTA (1,4,7-triazacyclononane-1,4,7-triacetic acid) and NODA (1,4,7-triazacyclononane-1,4-diacetic acid).

[0180] Further suitable chelating agents (eg comprising DOTA and DFO) are disclosed in WO 2022/133537, incorporated herein by reference.

[0181] In certain embodiments, the girentuximab may be covalently coupled to the radioactive isotope ^{124}I . This isotope is a positron emitter that can be attached to antibodies e.g. as described by Larsson et al. (J. Nucl. Med. 33 (1992), 2020-2023) or US 5,185,142, the content of which is herein incorporated by reference.

[0182] In any embodiment, radiolabelling of a protein or antibody is accomplished by covalent iodination, particularly with the Iodogen Reagent (1,3,4,6-tetrachloro-3a,6a-diphenyl glycoluril). Iodogen labeling is a solid phase oxidative method that is similar to the Chloramine-T method, but is generally considered to be milder, since the reaction takes place on the surface of the oxidant, minimizing exposure of the substrate (Salacinski, P.R.P., et al., Anal.Biochem. 117:136 (1981)).

[0183] Chelators with radiometals and other halogenated radioisotopes may be bound to antibodies via one or more amino acid residues or reactive moieties in the protein/antibody, including but not limited to one or more lysine residues, tyrosine residues or thiol moieties.

[0184] In another example, the antibody may be conjugated to a bifunctional linker, for example, bromoacetyl, thiols, succinimide ester, TFP ester, a maleimide, or using any amine or thiol- modifying chemistry known in the art.

[0185] The skilled person will be familiar with standard methods for conjugating chelating agents to antibodies and derivatives or fragments thereof. In addition, the skilled person will be familiar with approaches for selecting a relevant chelating agent for pairing with a radiometal, for example as described in Chem. Soc. Rev., 2014,43, 260, incorporated herein by reference.

[0186] In any aspect or embodiment, the radiolabelled girentuximab may comprise any radioisotope suitable for treatment of tumour or cancer masses. The radioisotope may be for use in alpha (α) or beta (β) or gamma (γ) therapy.

[0187] Examples of suitable radioisotopes for use in therapeutic applications include: actinium-225 (^{225}Ac), astatine-211 (^{211}At), bismuth-212 and bismuth-213 (^{212}Bi , ^{213}Bi), copper-64 and copper-67 (^{64}Cu , ^{67}Cu), iodine -123, -124, -125 or -131 (^{123}I , ^{124}I , ^{125}I , ^{131}I)

(¹²³I), lead-212 (²¹²Pb), lutetium-177 (¹⁷⁷Lu), radium-223 and radium-224 (²²³Ra, ²²⁴Ra), rhenium-186 and rhenium-188 (¹⁸⁶Re and ¹⁸⁸Re), samarium-153 (¹⁵³Sm), scandium-47 (⁴⁷Sc), strontium-90 (⁹⁰Sr), terbium-149 and terbium-161 (¹⁴⁹Tb and ¹⁶¹Tb) and yttrium-90 (⁹⁰Y).

[0188] In any embodiment, the radiolabelled girentuximab antibody or antigen binding fragment thereof is ²²⁵Ac- or ¹⁷⁷Lu- girentuximab wherein the radioisotope is conjugated to the girentuximab antibody directly or indirectly.

Cancers to be treated

[0189] As used herein, the term “cancer” refers to a malignant growth or tumour resulting from an uncontrolled division of cells. The term “cancer” includes primary tumours and metastatic tumours.

[0190] The methods of the present invention find particular utility in the treatment of cancers which have not previously been identified as being treatable utilising an agent for binding to CAIX.

[0191] In any embodiment, the cancer being treated is breast cancer. High levels of CAIX have previously been reported in breast cancers and CAIX expression has also been reported to be associated with resistance to chemotherapy or an indicator of treatment success. These observations date back over several decades and prior to now, treatment using a radiolabelled CAIX-binding agent has not previously been reported for this patient group.

[0192] The breast cancer may be so-called “triple negative breast cancer” (TNBC), an aggressive, metastatic and drug-resistant form of breast cancer has limited therapeutic options, and which is negative for other biomarkers of breast cancer such as the Estrogen receptor (ER positive breast cancer), Progesterone receptor (PR positive breast cancer) (both so-called “hormone receptor positive” cancers) and human epidermal growth factor receptor 2 (HER2 positive breast cancer).

[0193] In any embodiment, the breast cancer may be hormone receptor positive breast cancer, such as ER positive, PR positive, or ER&PR positive. In any embodiment, the breast cancer may be positive for HER2 (including HER2 and hormone receptor positive breast cancer).

[0194] In certain embodiments, the cancer being treated is not breast cancer.

[0195] In any embodiment, the cancer to be treated is cervical cancer. The cervical cancer may be a squamous cell carcinoma, or an adenocarcinoma. In any embodiment, a subject for whom treatment of a cervical cancer may exhibit one or more symptoms of cervical cancer such as abnormal vaginal bleeding, including contact bleeding, or pelvic pain. A subject considered at risk of cervical cancer may have had a previous infection with HPV 16 or 18 strains, or have one or more genetic markers indicative of a risk of cervical cancer.

[0196] In any embodiment, the cancer to be treated is colorectal cancer. In any embodiment, a subject for whom treatment of a colorectal cancer may exhibit one or more symptoms of colorectal cancer such as persistent change in bowel habits, rectal bleeding or blood in stools, persistent abdominal discomfort, weakness or fatigue, and unexplained weight loss. A subject considered at risk of colorectal cancer may have a family history of the disease or have one or more genetic markers considered to be associated with increased risk of colorectal cancer or may previously have had intestinal polyps.

[0197] In any embodiment, the cancer to be treated is esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma). A subject for whom treatment of esophageal cancer may exhibit one or more symptoms selected from: dysphagia, unexplained weight loss, chest pain, pressure or burning, worsening indigestion or heartburn or coughing or hoarseness. A subject considered at risk of esophageal cancer may have a family history of the disease or have one or more genetic markers considered to be associated with increased risk of colorectal cancer, or may previously have been diagnosed with Barrett's esophagus.

[0198] In any embodiment, the cancer to be treated is gastric cancer (including gastric adenocarcinoma). A subject for whom treatment of gastric cancer may exhibit one or more symptoms selected from: dysphagia, stomach pain, feeling bloated after small amounts of food, loss of appetite, indigestion, nausea and vomiting, fatigue, blackened stools. A subject considered at risk of gastric cancer may have a family history of the disease or have one or more genetic markers considered to be associated with increased risk of gastric cancer.

[0199] In any embodiment, the cancer to be treated is glioblastoma multiforme. A subject for whom treatment of glioblastoma may exhibit one or more symptoms including vision, hearing, balance, coordination, strength and reflex symptoms, nausea, vomiting, seizures or other neurological symptoms.

[0200] In any embodiment, the cancer to be treated is head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma). A subject for whom treatment of head and neck cancer may exhibit one or more symptoms such as pain, swelling, hoarseness, sore throat, persistent cough, halitosis, unexplained weight loss. A subject considered at risk of head and neck cancer may have a family history of the disease, have one or more genetic markers considered to be associated with increased risk of head and neck cancer, may have had a previous infection with HPV or Epstein-Barr virus, a weakened immune system, poor oral hygiene including gum disease, smoking or chewing betel nut, areca nut, gutka or pan, or have an inherited condition such as Fanconi anaemia or Li-Fraumeni syndrome.

[0201] In any embodiment, the cancer to be treated is liver cancer (including cholangiocarcinoma and hepatocellular carcinoma). As used herein, cholangiocarcinoma refers to biliary, or bile duct cancer. The cholangiocarcinoma may be intrahepatic, hilar or distal cholangiocarcinoma. The cancer may be gallbladder cancer or cancer of the ampulla of Vater. In any embodiment, a subject for whom treatment of a liver cancer (including cholangiocarcinoma and hepatocellular carcinoma) may be a subject exhibiting one or more symptoms of liver cancer (including cholangiocarcinoma and hepatocellular carcinoma).

[0202] As used herein, one or more symptoms of cholangiocarcinoma include: abdominal pain, yellowish skin (jaundice), weight loss, generalised itching, fever, light coloured stool or dark urine. The skilled person will be familiar with various risk factors for cholangiocarcinoma include primary sclerosing cholangitis (an inflammatory disease of the bile ducts), ulcerative colitis, cirrhosis, hepatitis C, hepatitis B, infection with certain liver flukes, and some congenital liver malformations. However, most people have no identifiable risk factors.

[0203] In any embodiment, the cancer to be treated is lung cancer (including non-small cell and small cell carcinoma). As used herein, the term "lung cancer" includes, but is not limited to all types of lung cancers at all stages of progression like lung carcinomas

metastatic lung cancer, non-small cell lung carcinomas (NSCLC) such as lung adenocarcinoma, squamous cell carcinoma, or small cell lung carcinomas (SCLC). In some embodiments, the subject suffers from a non-small cell lung carcinomas (NSCLC).

[0204] In any embodiment, the cancer to be treated is ovarian cancer (including epithelial ovarian carcinoma).

[0205] In any embodiment, the cancer to be treated is pancreatic cancer (including pancreatic ductal adenocarcinoma).

[0206] In any embodiment, the cancer to be treated is soft tissue sarcoma.

[0207] In particularly preferred embodiments, the cancer to be treated is not a renal cancer (including clear cell renal cancer).

[0208] In any embodiment, the cancer being treated is bladder cancer. The bladder cancer may be non-muscle-invasive bladder cancer (NMIBC).

Administration of radiolabelled agent (such as girentuximab) and methods of treatment

[0209] Methods for preparing an antigen binding protein into a suitable form for administration to a subject (e.g. a pharmaceutical composition) are known in the art and include, for example, methods as described in Remington's Pharmaceutical Sciences (18th ed., Mack Publishing Co., Easton, Pa., 1990) and U.S. Pharmacopeia: National Formulary (Mack Publishing Company, Easton, Pa., 1984).

[0210] The radiolabelled girentuximab antibody is usually administered as a pharmaceutical composition with a pharmaceutically acceptable carrier, e.g. physiological saline solution, or other aqueous carrier, optionally comprising a protein stabilizer such as human serum albumin (HSA). The compositions may contain pharmaceutically acceptable auxiliary substances as required to approximate physiological conditions such as pH adjusting and buffering agents, toxicity adjusting agents and the like, for example, sodium acetate, sodium chloride, potassium chloride, calcium chloride, sodium lactate and the like. The concentration of an antigen binding protein according to the present invention, in these formulations can vary widely, and will be selected primarily based on fluid volumes, viscosities, body weight and the like in accordance with the particular mode of administration selected and the patient's needs. Exemplary carriers include water,

saline, Ringer's solution, dextrose solution, and 5% human serum albumin. Non-aqueous vehicles such as mixed oils and ethyl oleate may also be used. Liposomes may also be used as carriers. The vehicles may contain minor amounts of additives that enhance isotonicity and chemical stability, e.g., buffers and preservatives.

[0211] Administering refers to the physical introduction of a composition comprising a therapeutic agent to a subject, using any of the various methods and delivery systems known to those skilled in the art including those described herein. Pharmaceutical compositions may be formulated from active agents of the invention as described herein for any appropriate route of administration.

[0212] The radiolabelled girentuximab is preferably administered intravenously, preferably by infusion or intravenous injections. The administration of the antibody by infusion is preferably performed over a period of up to about 30 minutes, more preferably in about 15 minutes. Of course, the radiolabelled girentuximab can also be administered by subcutaneous, intramuscular, intraperitoneal, intrathecal, intraventricular, intrasternal, and intracranial injection or infusion techniques.

[0213] The phrase 'therapeutically effective amount' or 'effective amount' generally refers to an amount of a radiolabelled girentuximab or antigen binding fragment thereof that (i) treats the particular disease, condition, or disorder, (ii) attenuates, ameliorates, or eliminates one or more symptoms of the particular disease, condition, or disorder, or (iii) delays the onset of one or more symptoms of the particular disease, condition, or disorder described herein. Undesirable effects, e.g. side effects, are sometimes manifested along with the desired therapeutic effect; hence, a practitioner balances the potential benefits against the potential risks in determining what is an appropriate "effective amount".

[0214] For instance, for the treatment of tumours, a therapeutically effective amount of the radiolabelled antibodies or compositions described herein may inhibit tumour growth by at least about 10%, by at least about 20%, by at least about 30%, by at least about 40%, by at least about 50%, by at least about 60%, by at least about 70%, by at least about 80%, or by at least about 90% or more, relative to untreated subjects. Alternatively, the treatments described herein may cause complete regression of the tumour mass. In other embodiments of the invention, tumour regression can be observed and continue for a period of at least about 10 days, at least about 20 days, at least about 30 days, at least

about 40 days, at least about 50 days or at least about 60 days, at least about 70 days, at least about 80 days, at least about 90 days, at least about 100 days or longer.

[0215] A therapeutically effective amount of a drug may also include a “preventative” or “prophylactically effective amount,” which is any amount of the radiolabelled girentuximab or antigen binding fragment thereof administered to a subject at risk of developing a cancer (eg a subject having a pre-malignant condition) or of suffering a recurrence of cancer, that inhibits the development or recurrence of the cancer. In certain embodiments, the prophylactically effective amount prevents the development or recurrence of the cancer entirely. “Inhibiting” or “preventing” the development or recurrence of a cancer means either lessening the likelihood of the cancer's development or recurrence, or preventing the development or recurrence of the cancer entirely.

[0216] The terms “treatment” or “treating” of a subject includes the application or administration of a compound of the invention to a subject with the purpose of delaying, slowing, stabilizing, curing, healing, alleviating, relieving, altering, remedying, less worsening, ameliorating, improving, or affecting the disease or condition, the symptom of the disease or condition, or the risk of (or susceptibility to) the disease or condition. The term “treating” refers to any indication of success in the treatment or amelioration of an injury, pathology or condition, including any objective or subjective parameter such as abatement; remission; lessening of the rate of worsening; lessening severity of the disease; stabilization, diminishing of symptoms or making the injury, pathology or condition more tolerable to the subject; slowing in the rate of degeneration or decline; making the final point of degeneration less debilitating; or improving a subject's physical or mental well-being.

[0217] As used herein, minimising or preventing the progression of cancer means treating the subject so as to prevent or delay the recurrence or metastasis of a tumour, or to prevent growth of an existing tumour. Minimising or preventing the progression of cancer includes preventing or delaying the recurrence of cancer, or preventing growth of an existing tumour, following treatment of cancer. The recurrence that is being prevented includes a recurrence for example, in the tumour bed, following surgical excision. Alternatively, recurrence includes metastasis of the cancer in another part of the body. The terms “preventing recurrence” and “preventing relapse” as used herein, are interchangeable.

[0218] The present invention also includes methods of preventing the development of cancer in an individual. For example, the individual for whom prevention of cancer is required may be considered to be at risk of developing cancer, but does not yet have detectable cancer. An individual at risk of the development of cancer may be an individual with a family history of cancer, and/or an individual for whom genetic testing or other testing indicates a high risk or high likelihood of the development of cancer. The individual may have cancer stem cells but does not yet have any detectable tumours. It will be understood that methods of preventing the development of cancer include methods of delaying the onset of cancer in a subject.

[0219] In any aspect or embodiment herein, the radiolabelled girentuximab may be administered in combination with (ie sequentially, simultaneously or following) administration of another treatment for the cancer. In any embodiment, the treatment may be any treatment for cancer, optionally selected from: surgery, immunotherapy, external beam radiation, chemotherapy or radio-immunotherapy.

[0220] Chemotherapeutic agents are chemical agents or drugs that are selectively destructive to cancer cells and tissues. Chemotherapeutics may include but are not limited to compounds such as, taxane compounds, compounds that act via taxane mechanisms, platinum compounds, anthracycline compounds, antimetabolites, epipodophyllotoxin compounds, camptothecin compounds, or any combination thereof.

[0221] In any aspect of the invention, the radiolabelled girentuximab and the additional treatment for cancer may be administered at the same time. Alternatively, they may be administered sequentially. For instance, the additional therapeutic (eg chemotherapeutic) may be administered prior to the radiolabelled girentuximab or the radiolabelled girentuximab may be administered prior to the additional therapeutic (eg chemotherapeutic). Alternatively, treatment with the additional therapeutic (eg chemotherapeutic) and/or the radiolabelled girentuximab may be staggered.

[0222] Suitable dosages of radiolabelled girentuximab or antigen binding fragment thereof for use according to the present invention will vary depending on the condition to be treated and/or the subject being treated. It is within the ability of a skilled physician to determine a suitable dosage, e.g., by commencing with a sub-optimal dosage and incrementally modifying the dosage to determine an optimal or useful dosage. Alternatively, to determine an appropriate dosage for treatment/prophylaxis, data from

the cell culture assays or animal studies are used, wherein a suitable dose is within a range of circulating concentrations that include the ED₅₀ of the active compound with little or no toxicity. The dosage may vary within this range depending upon the dosage form employed and the route of administration utilized. A therapeutically/prophylactically effective dose can be estimated initially from cell culture assays. A dose may be formulated in animal models to achieve a circulating plasma concentration range that includes the IC₅₀ (i.e., the concentration or amount of the compound which achieves a half-maximal inhibition of symptoms) as determined in cell culture. Such information can be used to more accurately determine useful doses in humans. Levels in plasma may be measured, for example, by high performance liquid chromatography.

[0223] The skilled person will appreciate that dosage of the agent for use in accordance with the methods of the invention will depend on various factors including the age, sex, height and weight of the subject to whom the agent is administered, and depending on the agent.

[0224] Preferably the girentuximab is administered to a subject or infused at a dose from about 1 mg to about 50 mg, preferably in a dose from about 5 mg to about 20 mg, and more preferably in a dose of about 10 mg. The specific activity of the radiolabeled antibody is preferably about 15 to about 20 MBq/mg, more preferably about 18 to about 19 MBq/mg.

[0225] In certain embodiments, the radiolabelled girentuximab is administered at a mass dose of about 5 to 20 mg of girentuximab by slow infusion.

[0226] In some embodiments an effective amount of antibody or antigen binding fragment thereof is administered to the patient identified as having a cancer at a loading dose of about 15 mg/kg, or about 20 mg/kg, or about 25 mg/kg on each of day 1 and day 8 of a first 21 -day cycle or on each of day 1 and day 8 of a first 28-day cycle, followed by administering a standard dose of the antibody or antigen binding fragment thereof to the patient, at about 15 mg/kg, about 20 mg/kg, or about 25 mg/kg on each of day 1 and day 8 of a subsequent 21 -day cycle or on each of day 1 and day 8 of a subsequent 28-day cycle.

[0227] In some embodiments, the radiolabelled girentuximab or antigen binding fragment thereof disclosed herein, is continued for a minimum duration of at least about

1 month, at least about 2 months, at least about 3 months, at least about 4 months, at least about 5 months, at least about 6 months, at least about 7 months, at least about 8 months, at least about 9 months, at least about 10 months, at least about 11 months, at least about 1 year, at least about 18 months, at least about 24 months, at least about 3 years, at least about 5 years, or at least about 10 years. In some embodiments, treatment with a radiolabelled girentuximab or antigen binding fragment thereof disclosed herein, is continued for a maximum duration of up to about 18 months, 16 months, 14 months, 12 months, 10 months, 8 months, or 6 months. The treatment duration may be from any of these minimum durations to any of these maximum durations, for example from 1 to 18 months.

[0228] As used herein, the terms “subject” “individual” and “patient” will be understood to be interchangeable. Although the invention finds application in humans, the invention is also useful for therapeutic veterinary purposes. The invention is useful for domestic or farm animals such as cattle, sheep, horses and poultry; for companion animals such as cats and dogs; and for zoo animals.

Kits

[0229] In another embodiment there is provided a kit or article of manufacture including a radiolabelled girentuximab or antigen binding fragment, as described above, preferably provided for use in a method or use outlined herein.

[0230] Optionally the kit further comprises a label or package insert with instructions for use.

[0231] The kit or “article of manufacture” may comprise a container and a label or package insert on or associated with the container. Suitable containers include, for example, bottles, vials, syringes, blister pack, etc. The containers may be formed from a variety of materials such as glass or plastic. The container holds a therapeutic composition which is effective for treating the condition and may have a sterile access port (for example the container may be an intravenous solution bag or a vial having a stopper pierceable by a hypodermic injection needle). The label or package insert indicates that the therapeutic composition is used for treating the condition of choice. In one embodiment, the label or package insert includes instructions for use.

[0232] The kit may comprise (a) a therapeutic composition; and (b) a second container with a second active principle or ingredient contained therein. The kit in this embodiment of the invention may further comprise a package insert indicating that the and other active principle can be used to treat a disorder or prevent a complication stemming from cancer. Alternatively, or additionally, the kit may further comprise a second (or third) container comprising a pharmaceutically-acceptable buffer, such as bacteriostatic water for injection (BWFI), phosphate-buffered saline, Ringer's solution and dextrose solution. It may further include other materials desirable from a commercial and user standpoint, including other buffers, diluents, filters, needles, and syringes.

[0233] In certain embodiments the therapeutic composition may be provided in the form of a device, disposable or reusable, including a receptacle for holding the therapeutic composition. In one embodiment, the device is a syringe. The device may hold 1-2 mL of the therapeutic composition. The therapeutic composition may be provided in the device in a state that is ready for use or in a state requiring mixing or addition of further components.

[0234] It will be understood that the invention disclosed and defined in this specification extends to all alternative combinations of two or more of the individual features mentioned or evident from the text or drawings. All of these different combinations constitute various alternative aspects of the invention.

[0235] It will be understood that the invention disclosed and defined in this specification extends to all alternative combinations of two or more of the individual features mentioned or evident from the text or drawings. All of these different combinations constitute various alternative aspects of the invention.

[0236] The examples that follow are intended to illustrate but in no way limit the present invention.

Examples

Example 1: Preparation of radiolabelled girentuximab

[0237] Radiolabelled girentuximab was prepared as previously described (see for example, WO 2021/000017). Briefly, bioconjugated girentuximab was prepared using

standard techniques to obtain DOTA-girentuximab, in ammonium acetate (pH 6), prior to being labelled with a radioisotope useful for therapeutic use (eg ^{177}Lu).

Example 2: *In vitro* and *in vivo* binding of radiolabelled girentuximab for targeting various cancers

[0238] Firstly, the ability of radiolabelled-DOTA-Girentuximab to bind to various cell lines was assessed. These data (shown in Figure 1A) demonstrate the ability of the antibody to bind to various cell types which express CAIX, although the degree of *in vitro* binding is variable. Negligible binding was observed for FaDu and HEK293 cells.

[0239] In a subsequent study, three groups of mice, each bearing different tumour xenografts, were tested. The groups were as follows:

Group 1: mice bearing an AsPc-1 cell xenograft (pancreatic cancer line); n= 4

Group 2: mice bearing a FaDu cell xenograft (squamous carcinoma pharynx/hypopharyngeal cancer cell line); n = 4

Group 3: mice bearing an HT-29 xenograft (colorectal cancer cell line); n = 4

[0240] The radiolabelled girentuximab was administered intravenously and imaging was conducted at 24 and 72 hour time points. Biodistribution was assessed at 72 hours following administration.

[0241] *Ex vivo* biodistribution of radiolabelled-DOTA-GmAb was compared to the *in vivo* biodistribution. Briefly, *ex vivo* biodistribution (shown in Figure 1B) corresponds to the distribution of radiolabelled-DOTA-GmAb in mouse organs, as assessed following necropsy. *In vivo* biodistribution corresponds to the biodistribution seen in whole-mouse imaging experiments.

[0242] Figure 1C shows that there was a high degree of correlation between *in vivo* and *ex vivo* quantification of the signal in the tumour.

Example 3: Efficacy study: radiolabelled girentuximab for targeting various cancers

[0243] Efficacy studies using ^{177}Lu -DOTA-Girentuximab were conducted to assess the ability of the radiolabelled antibody to treat non-RCC cancer types.

[0244] Six groups of mice, each bearing different tumour xenografts, were tested. The groups were as follows:

Group	No animals	Model	Treatment	Dose (MBq/inj)	Dose (µg/inj)
1	6	AsPc-1	Vehicle	N/A	N/A
2	6	AsPc-1	¹⁷⁷ Lu-DOTA-GmAb	10-12	60
3	6	FaDu	Vehicle	N/A	N/A
4	6	FaDu	¹⁷⁷ Lu-DOTA-GmAb	10-12	60
5	6	HT-29	Vehicle	N/A	N/A
6	6	HT-29	¹⁷⁷ Lu-DOTA-GmAb	10-12	60

[0245] The radiolabelled girentuximab was administered intravenously. The specific activity of the administered antibody was 200 MBq/mg.

[0246] Mice were followed up over a 10 week period to assess body weight and tumour volume.

[0247] Figure 2 shows changes in body weight in the mice over the treatment period. Overall, the results indicate that the treatment was well tolerated.

[0248] Figure 3 shows the changes in tumour volume in the mice over the treatment period.

[0249] Figure 4 shows survival curves from the different study groups.

[0250] Overall, the results show that radiolabelled girentuximab was effective in treating all three cancer types, as evidence by reduced tumour volume and increased survival, compared to animals receiving vehicle.

[0251] Interestingly, reduction in tumour volume was most apparent in mice bearing the FaDu xenograft, despite the fact that earlier *in vitro* data (Figure 1A) indicated poor binding of radiolabelled GmAb to this particular cell type. Without being bound by theory, the inventors believe that it is possible that this cell line has increased sensitivity to radiation and therefore can be treated with radiolabelled GmAb despite having only limited expression of CAIX. Tumour volume was also reduced in mice treated with radiolabelled GmAb and bearing the HT-29 or AsPc-1 xenografts.

[0252] Survival curves showed significant differences between treated and non-treated animals in all three xenograft models, although the effect was more pronounced for mice bearing the AsPc-1 and HT-29 xenografts.

[0253] The inventors also observed that the most rapidly growing cancer, HT-29, responded less to treatment, compared to the more slowly growing AsPc-1 cancer cells, despite HT-29 cells having greater uptake of the antibody (as assessed in biodistribution studies). Without wishing to be bound by theory, the inventors believe this discrepancy between degree of uptake and response to treatment, could be due to differences in intrinsic radiosensitivity between the cancer types.

[0254] Another surprising observation was that FaDu cells (which are understood in the art to be intrinsically radiosensitive), and had the lowest uptake of all three cancer types, responded well to treatment. This is despite negligible *in vitro* binding to FaDu cells.

[0255] Overall these results indicate that *in vitro* binding of a radiolabelled antibody and uptake of radiolabelled antibody cannot be relied upon as an indicator of likely treatment success.

Example 4: Treatment of alternative cancer types

[0256] Similar experiments to those conducted in Example 3 are performed using ¹⁷⁷Lu-DOTA-GmAb to treat tumour xenografts in mice as follows:

Group 1: mice bearing an A-549 cell line xenograft (lung cancer)

Group 2: mice bearing a MDA-MB-468 cell line xenograft (triple negative breast cancer)

Group 3: mice bearing a HeLa cell line xenograft (cervical cancer)

Group 4: mice bearing a AGS cell line xenograft (gastric cancer)

Group 5: mice bearing a HepG2 cell line xenograft (liver cancer)

Group 6: mice bearing a A2780 cell line xenograft (ovarian cancer)

Group 7: mice bearing a SK-LMS1 cell line xenograft (soft tissue sarcoma – vulvar leiomyosarcoma)

[0257] At 10 days following intravenous injection of ^{177}Lu -DOTA-GmAb into the xenograft-bearing mice, the mice are assessed to determine efficacy of therapeutic intervention with the radiolabelled antibody.

[0258] The results will show that the radiolabelled antibody is able to bind to the tumour xenografts and will result in reduction in tumour volume, and increased survival, compared to mice bearing the same tumour xenografts and receiving treatment with vehicle only.

[0259] These results will indicate that radiolabelled-GmAb is suitable for use in treatment of various cancer types.

Example 5: Treatment of breast cancer

[0260] Triple Negative Breast Cancer (TNBC) is an aggressive, metastatic and drug-resistant cancer with limited therapeutic options.

[0261] The inventors believe that CAIX, a hypoxia-mediated breast tumour growth regulator, is important for the maintenance of breast cancer stem cells within hypoxic regions of breast cancer tissue. Accordingly, the inventors firstly assessed imaging of TNBC using PET/CT imaging with ^{89}Zr -labeled girentuximab in 12 metastatic TNBC patients.

[0262] Patients underwent imaging with Fludeoxyglucose F18 (FDG) and ^{89}Zr -girentuximab PET-CT and CT. Patients received a single slow intravenous administration of $37 \pm 10\%$ MBq ^{89}Zr -girentuximab (10 mg). At day 3 post administration, a skull to mid-thigh PET/CT was acquired with 10 minute acquisition time per bed position. The gold standard was determined by FDG PET/CT, CT and follow-up; lesion detected at least by 2 modalities was considered as true positive. Tumour $\text{SUV}_{[\text{max}, \text{mean}]}$, total lesion glycolysis (TLG) and metabolic tumour volume (MTV) were measured. Immunohistochemistry (IHC) was performed with Bond RX fully automated research staining with anti-CAIX antibody

(Leica, clone TH22). Staining was evaluated with semi-quantitative analysis (percentage and intensity of tumour cells expression) and SUV values compared to the degree of CAIX expression evaluated by IHC.

[0263] Preliminary results from 4 patients were examined and include data derived from a total of 49 lesions (lymph nodes, bone, lung and breast) which were detected in these patients (41 by ^{89}Zr -girentuximab, 42 by CT and 49 by FDG PET/CT). Forty-four lesions were confirmed by gold standard: 24, 5, 4, 2, 9 in nodes, lung, bone, skin and breast respectively.

[0264] Overall sensitivity of ^{89}Zr -girentuximab PET/CT was 93.2%, with 100% sensitivity for bone, lung, breast, skin and 87.5% for nodes. Overall sensitivity for both CT and FDG-PET/CT was 82.7%. Median tumor SUV_{max} were 3.45 [IQ: 2.03-4.69] and 4.68 [IQ: 3.27-10.71] for ^{89}Zr -girentuximab and ^{89}Zr -girentuximab FDG, respectively. IHC showed two CAIX-high-expression lesions [100%, 20%] for two patients whereas two patients presented respective low profiles [3%, 0%]. IHC CAIX cell status and ^{89}Zr -girentuximab SUV_{mean} presented a weak correlation ($\rho=0.80$; $p=0.20$). No ^{89}Zr -girentuximab safety issues were reported.

[0265] The results demonstrate that ^{89}Zr -girentuximab is useful for PET/CT imaging and diagnosis of TNBC in patients and provides a superior result to biopsy IHC.

[0266] The inventors next test whether radio-labelled girentuximab successfully treats TNBC.

[0267] A therapeutic efficacy study is conducted in subjects having TNBC. The therapeutic agent used is ^{177}Lu -DOTA-GmAb. The results will show that radiolabelled-girentuximab successfully treats TNBC.

Example 6: Treatment of bladder cancer

[0268] Patients with non-muscle-invasive bladder carcinoma (NMIBC) are usually treated with cystectomy. Therefore, new treatment options which enable preservation of the urinary bladder are needed.

[0269] CAIX is expressed on the luminal surface of papillary structures in direct contact with vesical cavity. The inventors conducted a pilot prospective study to determine the feasibility of radioimmunotherapy using intravesically instilled girentuximab antibody

labelled with astatine-211. This pilot prospective study was aimed firstly at ensuring intravesical radioactivity confinement and tumour targeting after intravesical instillation of ^{89}Zr -girentuximab.

[0270] Patients had one intravesical instillation of $37 \pm 10\%$ MBq of ^{89}Zr -girentuximab (10 mg) and retained urine for 2 hours. Then 4 PET/CT scans were performed, 3 with one step on pelvis (H+2, Day1 and D2) and one from skull to mid-thigh at H+4 to observe intravesical radioactivity evolution over time.

[0271] A blood sample was taken at D1 to quantify a possible vascular passage of radioactivity. For all acquisitions, 10 minutes acquisition time per bed position was used. The gold standard was determined by a second look cystoscopy and trans-urethral resection bladder (TURB) on ^{89}Zr -girentuximab PET/CT positive sites. Immunohistochemistry (IHC) was performed with an anti-CAIX antibody (Leica, clone TH22). Staining was evaluated with semi-quantitative analysis (percentage and intensity of tumour cells expression) and ^{89}Zr -girentuximab PET/CT bladder pattern compared to the degree of CAIX expression evaluated by IHC.

[0272] Results from 4/6 patients were obtained. Despite previous multiple previous intravesical instillations with bacillus Calmette-Guerin (BCG – a commonly used intravesical immunotherapy to treat bladder cancer), recurrent pTaG3 was identified in each patient.

[0273] ^{89}Zr -girentuximab PET/CT showed no extra-vesical leakage. In 2/4 patients with positive IHC, uptake spots on the bladder wall was confirmed by TURB for one patient with corresponding recurrence foci and inflammatory scarring reaction for the second one. For the two other patients no uptake was observed in line with negative IHC. No adverse radiation contamination was observed during the process and no particular worker exposure was observed.

[0274] The results (as shown in Figure 5 and 6) indicate that intravesical instillation of ^{89}Zr -girentuximab showed radioactivity confinement into the bladder and, in patients with positive IHC, indicating that ^{89}Zr -girentuximab is useful for detecting and imaging tumours in this patient group.

[0275] A therapeutic efficacy study is next conducted in subjects having bladder cancer and where the therapeutic agent used is ^{98}Lu - girentuximab. The results will show that radiolabelled- girentuximab successfully treats bladder cancer.

CLAIMS

1. A method of treating, preventing or minimising progression of cancer in a subject, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX, wherein the cancer is selected from:
 - bladder cancer
 - breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
 - cervical cancer
 - colorectal cancer
 - esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
 - gastric cancer (including gastric adenocarcinoma)
 - glioblastoma multiforme
 - head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
 - liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
 - lung cancer (including non-small cell and small cell carcinoma)
 - ovarian cancer (including epithelial ovarian carcinoma)
 - pancreatic cancer (including pancreatic ductal adenocarcinoma), and
 - soft tissue sarcoma

thereby treating, preventing or minimising progression of the cancer in the subject.

2. A method of minimising, reducing or preventing growth of a cancer tumour in a subject, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby minimising, reducing or preventing growth of the cancer tumour in the subject.

3. A method of minimising, reducing or preventing metastasis of cancer in a subject, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby minimising, reducing or preventing metastasis of the cancer in the subject.

4. A method of increasing survival of a subject suffering from cancer, the method comprising:

- administering to a subject in need thereof, a radiolabelled agent for binding to CAIX,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and
- soft tissue sarcoma

thereby increasing survival of the subject.

5. Use of a radiolabelled agent for binding to CAIX, in the manufacture of a medicament for:

- treating, preventing or minimising progression of cancer in a subject,
- minimising, reducing or preventing growth of a tumour in a subject,
- minimising, reducing or preventing metastasis in a subject, or
- increasing survival of a subject suffering from cancer,

wherein the cancer is selected from:

- bladder cancer
- breast cancer (including triple negative breast cancer, hormone receptor positive breast cancer (ER positive, PR positive, ER/PR positive), and HER2 positive breast cancer)
- cervical cancer
- colorectal cancer
- esophageal cancer (including esophageal squamous cell carcinoma (SCC) and esophageal/esophagogastric junction adenocarcinoma)
- gastric cancer (including gastric adenocarcinoma)
- glioblastoma multiforme
- head and neck cancer (including head and neck squamous cell carcinoma, hypopharyngeal and nasopharyngeal carcinoma)
- liver cancer (including cholangiocarcinoma and hepatocellular carcinoma)
- lung cancer (including non-small cell and small cell carcinoma)
- ovarian cancer (including epithelial ovarian carcinoma)
- pancreatic cancer (including pancreatic ductal adenocarcinoma), and

- soft tissue sarcoma.

6. The method of any one of claims 1 to 4, or the use of claim 5, wherein the radiolabelled agent for binding to CAIX is a radiolabelled small molecule, peptide or polypeptide.

7. The method or use of claim 6, wherein the radiolabelled agent for binding to CAIX is a radiolabelled small molecule, wherein the small molecule is selected from the group consisting of: SLC-0111, SLC-149, SLC-0121, SLC-101, PMI-05, sulfamide-nitroimidazole, JS-403, UB-TT220, HEHEHE-Z09781, -MIP-1486, MIP-1490, MIP-1504 (especially ^{99m}Tc-HEHEHE-Z09781, ^{99m}Tc-MIP-1486, ^{99m}Tc-MIP-1490 or ^{99m}Tc-MIP-1504/5) and PHC-102.

8. The method or use of claim 6, wherein the radiolabelled agent for binding to CAIX is a radiolabelled peptide, wherein the peptide is selected from the group consisting of: 3B-301, 3B-302 or CAIX-P1.

9. The method or use of claim 6, wherein the radiolabelled agent for binding to CAIX is radiolabelled antibody or radiolabelled antigen-binding fragment thereof.

10. The method or use of claim 9, wherein the radiolabelled agent is in the form of a radiolabelled antibody or radiolabelled antigen binding fragment thereof that comprises one or more Fc regions of an antibody.

11. The method or use of claims 9 or 10, wherein the antibody for binding to CAIX is selected from radiolabelled girentuxumab (G250), BCA-356, BAY-794620 or SLC-0131.

12. The method or use of claim 11, wherein the radiolabelled agent is radiolabelled girentuximab, or a functional variant or fragment thereof that retains the ability to bind to CAIX.

13. The method or use of claim 12, wherein the radiolabelled girentuximab is radiolabelled chimeric girentuxumab antibody (also referred to as cG250).

14. The method or use of claim 12, wherein the radiolabelled girentuximab is radiolabelled humanised girentuxumab antibody (also referred to as hG250).

15. The method or use of any one of the preceding claims, wherein the agent and radiolabel are conjugated directly or via a chelator or linker moiety.
16. The method or use of any one of the preceding claims, wherein the radiolabelled agent comprises a radiolabel selected from: actinium-225 (^{225}Ac), astatine-211 (^{211}At), bismuth-212 and bismuth-213 (^{212}Bi , ^{213}Bi), copper-64 and copper-67 (^{64}Cu , ^{67}Cu), iodine-123, -124, -125 or -131 (^{123}I , ^{124}I , ^{125}I , ^{131}I) (^{123}I), lead-212 (^{212}Pb), lutetium-177 (^{177}Lu), radium-223 and radium-224 (^{223}Ra , ^{224}Ra), rhenium-186 and rhenium-188 (^{186}Re and ^{188}Re), samarium-153 (^{153}Sm), and scandium-47 (^{47}Sc), strontium-90 (^{90}Sr), terbium-149 and terbium-161 (^{149}Tb and ^{161}Tb), and yttrium-90 (^{90}Y).
17. The method or use of claim 16, wherein the radiolabel is lutetium-177 (^{177}Lu) or actinium-225 (^{225}Ac).
18. The method or use of any one of claims 12 to 15, wherein the radiolabelled agent is selected from ^{177}Lu -girentuximab or ^{225}Ac girentuximab.
19. The method or use of any one of claims 12 to 18, wherein the radiolabelled girentuximab comprises one or more amino acid substitutions in the Fc region of the antibody which reduce the serum half-life of the antibody.
20. The method or use of any one of claims 12 to 19, wherein the radiolabelled girentuximab is administered as the sole treatment for a cancer.
21. The method or use of any one of claims 12 to 19, wherein the radiolabelled girentuximab is administered subsequent to a previous treatment for cancer (optionally wherein the previous treatment comprises surgery, chemotherapy, immunotherapy, external beam radiation, autologous stem cell therapy, or combinations thereof).
22. The method or use of any one of the preceding claims wherein the cancer is pre-cancerous or non-metastatic.
23. The method or use of any one of claims 1 to 21, wherein the cancer is malignant or metastatic.
24. The method or use of any one of the preceding claims wherein the method or use further comprises identifying a subject in need of treatment.

25. The method or use of any one of the preceding claims wherein the method or use further comprises determining whether the subject has a cancer expressing or overexpressing CAIX.
26. The method or use of any one claims 13 to 26, wherein the cancer is cervical cancer.
27. The method or use of any one of claims 13 to 26, wherein the cancer is colorectal cancer.
28. The method or use of any one of claims 13 to 26, wherein the cancer is esophageal cancer.
29. The method or use of any one of claims 13 to 26, wherein the cancer is gastric cancer.
30. The method or use of any one of claims 13 to 26, wherein the cancer is glioblastoma multiforme.
31. The method or use of any one of claims 13 to 26, wherein the cancer is head and neck cancer (such as hypopharyngeal cancer, pharyngeal cancer).
32. The method or use of any one of claims 13 to 26, wherein the cancer is liver cancer.
33. The method or use of any one of claims 13 to 26, wherein the cancer is lung cancer.
34. The method or use of any one of claims 13 to 26, wherein the cancer is ovarian cancer.
35. The method or use of any one of claims 13 to 26, wherein the cancer is pancreatic cancer.
36. The method or use of any one of claims 13 to 26, wherein the cancer is soft tissue sarcoma.

37. The method or use of any one of claims 13 to 26, wherein the cancer is breast cancer (including breast cancer selected from: triple negative breast cancer, hormone receptor positive breast cancer and HER2 positive breast cancer).

38. The method or use of any one of claims 13 to 26, wherein the cancer is bladder cancer.