

Methods for the diagnosis and/or treatment of prostate cancer

Field

[0001] The present invention relates generally to methods for diagnosis by radioimaging and radiotherapy comprising the administration of specific compounds complexed with a radioisotope to deliver a dose of radiation to a specific site for the imaging or treatment of a prostate cancer.

Background

[0002] Prostate cancer is the second most frequent malignancy in men worldwide and is the second most common cancer, accounting for 9.5% of all new cancers in 2018. The occurrence of prostate cancer varies and correlates with age, with incidence between 30% of males aged 40 to 50 years old and 50% to 80% of males aged 80 and over. At initial presentation, 80% of patients have local disease, 12% have regional disease and 4% have metastatic disease. Although the 5-year survival rate for patients with local or regional prostate cancer is 99%, the survival rate drops to about 30% in the case of metastatic disease.

[0003] Prostate-specific membrane antigen (PSMA) is a type II transmembrane glycoprotein that is expressed in normal, benign and malignant prostate tissues. The expression of the PSMA membrane protein increases with aggressiveness of the prostate tumour, the presence of metastatic disease and recurrence of the cancer. For example, expression of the PSMA membrane protein is 100- to 1000-fold higher in prostatic adenocarcinoma than in benign prostate tissue and increases with androgen deprivation, with levels of the protein highest in high-grade and castration resistant prostate cancer. While the level of PSMA expression is correlated with development of disease, there are cases of prostate cancer where increased expression of PSMA is not observed in biopsies.

[0004] While the PSMA membrane protein is expressed in normal prostate tissue, with an increase in expression in cancerous tissue, other healthy tissues also express the membrane protein, for example, salivary glands, duodenal mucosa, proximal renal tubular cells and some neuroendocrine cells in the colonic crypts. Even if treatments specifically targeting the PSMA membrane protein are developed, the presence of the protein on healthy tissues often means that unwanted off-site damage can occur.

[0005] Current techniques for the treatment of prostate cancer includes the use of a ^{99m}Tc radioisotope with a combination of MRI and CT imaging, however this approach has limited sensitivity and specificity for prostate cancer. The use of ^{18}F -FDG with PET imaging provides some improvement, however the low sensitivity of this technique limits its use for the diagnosis of prostate cancer.

[0006] The use of a radioisotope to diagnose and treat prostate cancer requires the use of a ligand that can coordinate the desired radioisotope and target the selected site. Urea-based ligands are known to target the PSMA membrane protein, however no such ligands have been approved for use by the FDA. While ^{177}Lu -PSMA has shown some promise, however toxicity to the patient and unwanted radiation damage to healthy tissue have also been observed.

[0007] There remains a need for regimes to diagnose and/or treat cancers associated with expression of the PSMA membrane protein, specifically prostate cancer, where the regime is tolerated by the patient and off-target effects are limited.

Summary of the invention

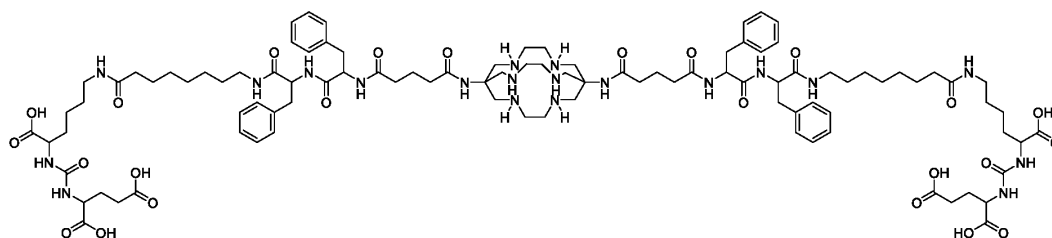
[0008] The present invention provides a method for radioimaging and/or treating a cancer associated with overexpression of a PSMA membrane antigen by administration of a compound of Formula (I), or a salt thereof, complexed with a suitable radioisotope to a subject in need thereof. The compound of Formula (I) is symmetrical and comprises a sarcophagine fragment that is capable of complexing copper (Cu) ions and two urea fragments that are known to bind to prostate-specific membrane antigen (PSMA), a type II transmembrane glycoprotein. Each urea fragment is bound to the sarcophagine by the same linker, resulting in a symmetrical molecule. The urea fragment comprises lysine and glutamate residues, where the lysine fragment in each urea is attached to the linker. Where the compound of Formula (I) is complexed with a ^{64}Cu radioisotope and administered to a subject, radioimaging by positron emission tomography (PET) reveals where the radiolabelled complex is localised. And where the compound of Formula (I) is complexed with a ^{67}Cu radioisotope and administered to a subject, therapeutic effects in the treatment of cancers associated with expression of the PSMA membrane protein may be realised. Since the urea fragments of Formula (I) bind to PSMA, the localisation of the compound signals an area where the PSMA membrane protein is overexpressed likely indicates the presence of a cancer. Without being bound by theory, the

present inventors believe that the presence of two groups capable of binding to the PSMA membrane protein in the compound of Formula (I) leads to greater affinity for the membrane protein and therefore better binding. This having benefits in both imaging cancers and treating said same cancers.

[0009] Although PSMA is expressed in normal, benign and malignant prostate tissue, expression of the membrane protein increases with tumor aggressiveness, metastatic disease and recurrence. For example, PSMA is expressed 100- to 1000-fold higher in prostatic adenocarcinoma than in the benign prostate.

[0010] Since PSMA is also expressed in healthy tissue (e.g. salivary glands, duodenal mucosa, proximal renal tubular cells and subpopulations of neuroendocrine cells in the colonic crypts), administration of a radiolabelled compound that attempts to target PSMA often results in unwanted off-site effects and damage of healthy tissue.

[0011] The present inventors have found that the administration of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a copper radioisotope allows for the delivery of a high dose of radiation to a cancer site where the PSMA membrane protein is overexpressed:

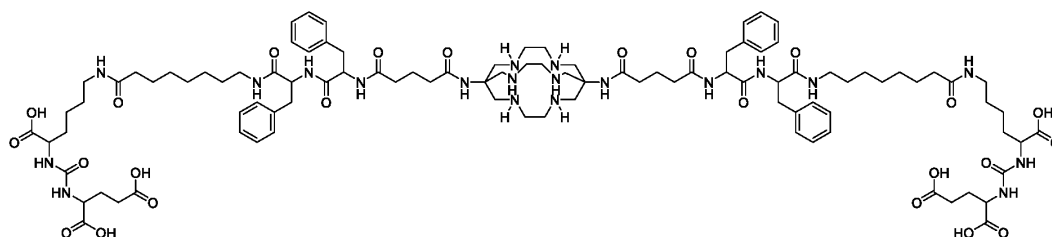


Formula (I)

[0012] Since the compound of Formula (I) shows specificity for the PSMA membrane protein, binding of the compound containing the copper radioisotope at other sites is reduced. This in turn reduces the severity and incidence of adverse events in patients, which means the treatment has higher tolerability. Furthermore, the present inventors have found that since the compound of Formula (I) shows better retention of the copper radioisotope and better binding at the target site, the amount of the radiolabelled compound that is administered may be reduced. The

present inventors believe that the radiolabelled compounds of Formula (I) may be used for radioimaging and radiotherapeutic treatments of cancers where the PSMA membrane protein is overexpressed, since the compound of Formula (I) will bind and localise at the sites where the protein is expressed. Imaging of the subject by PET, SPECT, and/or CT after administration of the ^{67}Cu -labelled compound of Formula (I) may be performed to confirm the localisation of the radiolabelled compound and targeted treatment. This could be achieved using the same bis-compound but radiolabelled with ^{64}Cu . Where a compound of Formula (I) labelled with a copper radioisotope is administered and subsequent imaging is performed, this allows for the location of the radiolabelled compound and hence the cancer associated with expression of the PSMA membrane protein to be determined. The PSMA membrane protein is expressed in normal tissue, with cancerous tissue expressing comparatively higher levels of the PSMA membrane protein such that PSMA is "overexpressed", i.e. in higher concentrations than in normal tissue.

[0013] In an aspect, the present invention provides a method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:

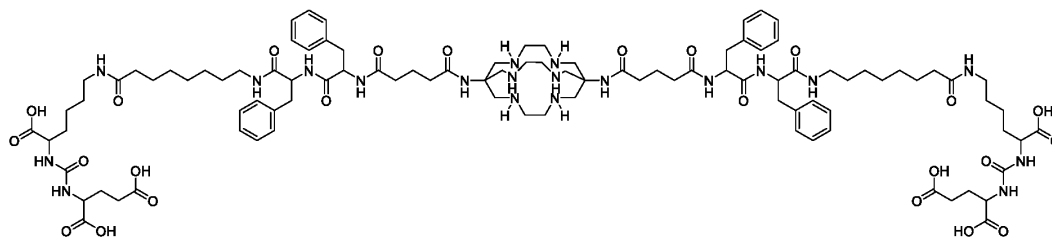


Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer and/or mediate a decrease in prostate specific antigen (PSA) levels.

[0014] In another aspect, the present invention provides a method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a

compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:

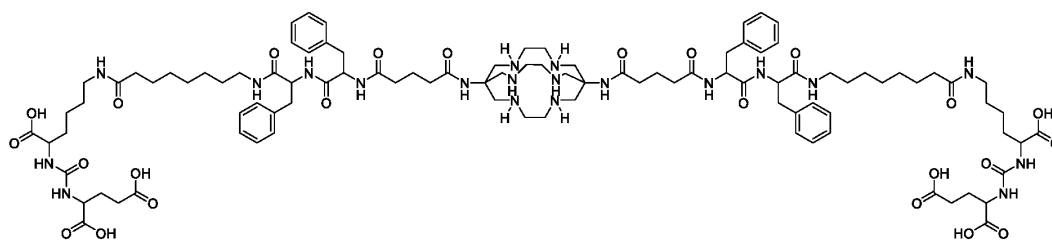


Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer.

[0015] In some embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is about 4 GBq, about 8 GBq, about 12 GBq, about 16 GBq, about 20 GBq or about 24 GBq. In some embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is more than about 12 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is more than about 16 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is more than about 20 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is the highest dose that is tolerated by the subject.

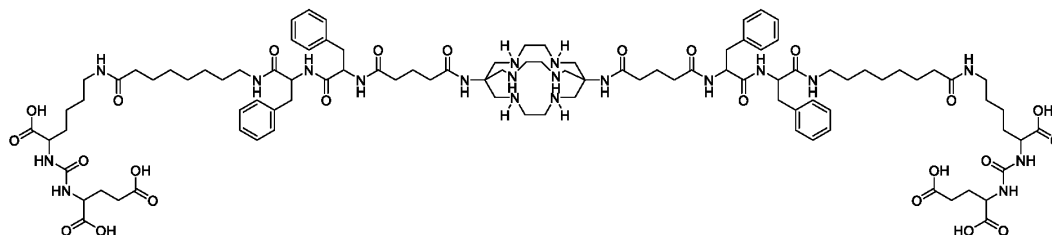
[0016] In certain embodiments, the present invention provides a method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:



Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 4 GBq.

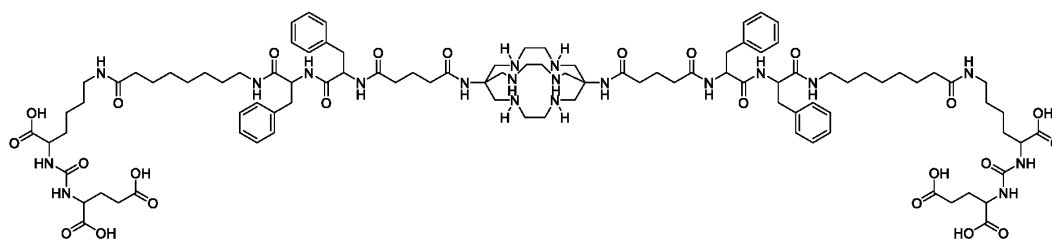
[0017] In some embodiments, the present invention provides a method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:



Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 8 GBq.

[0018] In some embodiments, the present invention provides a method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:



Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 12 GBq.

[0019] In an embodiment, the cancer is prostate cancer. In another embodiment, the cancer is metastatic prostate cancer. In a further embodiment, the prostate cancer is PSMA-expressing metastatic castrate resistant prostate cancer (mCRPC).

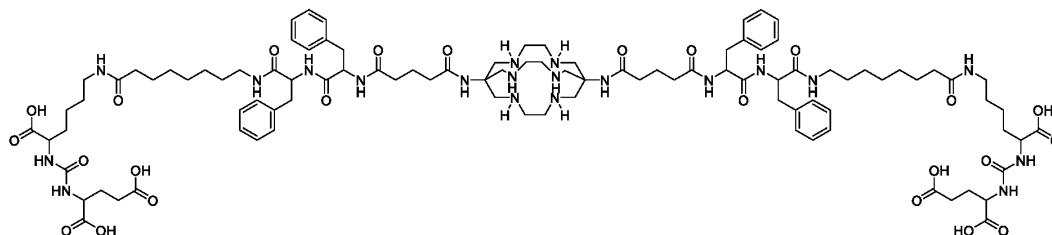
[0020] In a further embodiment, the prostate cancer is PSMA-expressing metastatic castrate resistant prostate cancer (mCRPC) is progressive mCRPC despite prior androgen deprivation therapy and at least either enzalutamide and/or abiraterone (or other such androgen receptor pathway inhibitors).

[0021] In a further embodiment the subject is a male subject with a castrate level of serum/plasma testosterone of about <50 ng/dL or about <1.7 nmol/L.

[0022] The present inventors have found that the administration of a compound of Formula (I) or a pharmaceutically acceptable salt thereof to a subject in need thereof can result in the reduction of a prostate specific antigen (PSA) level, where the compound of Formula (I) is complexed with a Cu radioisotope. Without wishing to be bound by theory, the present inventors believe that the administration of the compound of Formula (I) complexed with a Cu radioisotope leads to a reduction in the size of lesions associated with prostate cancer present in the subject. Exposure of the lesion to the radioactive decay products associated with the Cu radioisotope results in the reduction in size and/or volume of the lesion in the subject. This can be seen in Figures 5, 6, 7 or 8, where the size of the lesion is reduced after administration of the compound of Formula (I) complexed with a ^{67}Cu radioisotope. Furthermore, as seen in Figures 5, 6, 7 and 8, in certain embodiments, the lesion is not detectable by PET imaging after the administration of two doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope, i.e. after treatment with two cycles. Therefore in certain embodiments, the present invention provides a method for the treatment of a cancer, where the lesion associated with the cancer is not detected after administration of one or more doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope. In certain embodiments, the present invention provides a method for the treatment of a cancer, where the lesion associated with the cancer is not detected after administration of two doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope. In other embodiments, the present invention provides a method for the treatment of a cancer, where the lesion associated with the cancer is not detected after administration of three doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope. Given the nature of the lesion (i.e. being associated with prostate cancer), the

present inventors believe that the reduction in the size of the lesion as a result of the compound of Formula (I) then results in a reduction in the level of PSA in the subject.

[0023] Therefore, according to a further aspect, the present invention provides a method for reducing the prostate specific antigen (PSA) level in a subject, the method comprising administering to the subject in need thereof a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount:

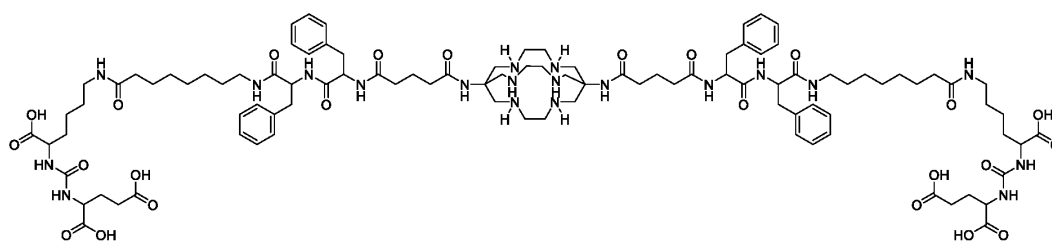


Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present.

[0024] In certain embodiments, the PSA level of the subject prior to administration is greater than about 0.1 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 1 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 10 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 100 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 500 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 1000 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 1500 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 2000 ng/ml.

[0025] In another aspect, the present invention provides a method for reducing the prostate specific antigen (PSA) level in a subject, the method comprising administering to the subject in need thereof a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount:



Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0 ng/ml, wherein the prostate gland of the subject is absent.

[0026] In some embodiments, the reduction in the PSA level of the subject at a time of about four weeks after administration of the compound of Formula (I) is between about 20% and about 80%, when compared to the PSA level of the subject prior to administration of the compound of Formula (I). In some embodiments, the PSA level of the subject at a time of about four weeks after administration of the compound of Formula (I) is reduced by about 20%, about 30%, about 40%, about 50%, about 60%, about 70% or about 80% when compared to the PSA level of the subject prior to administration of the compound of Formula (I). In other embodiments, the PSA level of the subject at a time of about four weeks after administration of the compound of Formula (I) is reduced by more than 80% when compared to the PSA level of the subject prior to administration of the compound of Formula (I). In other embodiments, the PSA level of the subject at a time of about four weeks after administration of the compound of Formula (I) is reduced by more than 90% when compared to the PSA level of the subject prior to administration of the compound of Formula (I). In other embodiments, the PSA level of the subject at a time of about four weeks after administration of the compound of Formula (I) is reduced by more than 95% when compared to the PSA level of the subject prior to administration of the compound of Formula (I). In other embodiments, the PSA level of the subject at a time of about four weeks after administration of the compound of Formula (I) is reduced by more than 99% when compared to the PSA level of the subject prior to administration of the compound of Formula (I).

[0027] In some embodiments, the method further comprises the administration of a second dose of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope, wherein the dose of radiation delivered by the second administration of the compound of Formula (I) is the same or different to the first dose of radiation delivered by

the first administration of the compound of Formula (I). In some embodiments, the dose of radiation delivered by the second administration of the compound of Formula (I) is the same as the dose of radiation delivered by the first administration of the compound of Formula (I).

[0028] Where a second dose of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope is to be administered, the second dose is administered about four weeks after the first dose. In certain embodiments, the second dose is administered at least four weeks after the first dose. In certain embodiments, the second dose is administered about six weeks after the first dose. In other embodiments, the second dose is administered about eight weeks after the first dose. In other embodiments, the second dose is administered about 12 weeks after the first dose. In some embodiments, the second dose is administered about 6 months, about 12 months, about 18 months, about 24 months, about 30 months or about 36 months after the first dose.

[0029] The present inventors have found that the PSA level of the subject is reduced after administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope. In some embodiments, the PSA level of the subject is reduced after a first administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope. In other embodiments, the PSA level of the subject is reduced after a second administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope. This can be seen in Figure 4, where administration of a single dose of the compound of Formula (I) complexed with a ^{67}Cu radioisotope resulted in a reduction in the PSA level of the subject, while a further administration of the compound resulted in undetectable levels of PSA.

[0030] In certain embodiments, the reduction in the PSA levels of the subject at a time of about 2 weeks after administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope is about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80% or about 90%, when compared to the PSA level of the subject prior to administration of the compound.

[0031] In certain embodiments, the reduction in the PSA level of the subject at a time of about 2 weeks after a second administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope is about 20%, about 30%, about 40%,

about 50%, about 60%, about 70%, about 80% or about 90%, when compared to the PSA level of the subject prior to administration of the compound.

[0032] The present inventors have found that the administration of at least one dose of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope leads to a reduction in the levels of PSA in the subject. Furthermore, the present inventors have found that after a time, PSA is not detected in the subject after administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof, wherein the limit of detection is about 0.01 ng/ml.

[0033] In certain embodiments, PSA is not detected in the subject after administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope. In other embodiments, PSA is not detected in the subject at a time of about four weeks after administration of the compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a Cu radioisotope.

[0034] In a further embodiment the subject is a male subject with a prostate specific antigen (PSA) level of > 2 ng/mL or above for more than 3 weeks before administering an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ⁶⁷Cu radioisotope. However, some individuals with PSA levels below 2.0 ng/mL may also have prostate cancer and be administered an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ⁶⁷Cu radioisotope.

[0035] In a further embodiment the subject experiences a decrease in percentage of PSA, alkaline phosphatase (ALP) and lactate dehydrogenase (LDA) biomarkers after one treatment administration cycle of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ⁶⁷Cu radioisotope relative to a baseline taken prior the treatment.

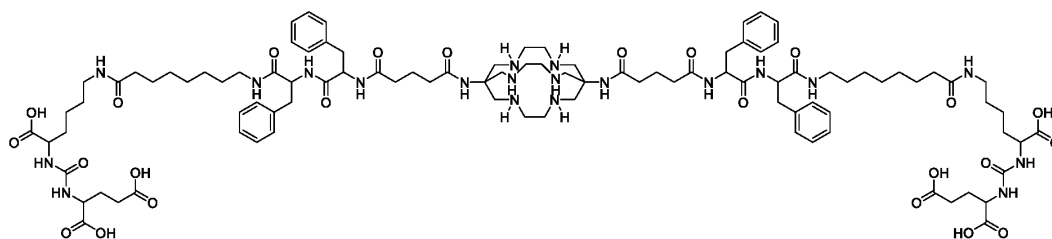
[0036] In a further embodiment the method comprises administering to a subject an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ⁶⁷Cu radioisotope with 1, 2, 3, or 4 treatment cycles.

[0037] In further embodiments the method comprises administering to a subject an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope, at least 2 administrations, about 6 to 14 weeks apart, of a dose to provide between 4 to 24 GBq by IV slow infusion over about 30 minutes to about 60 minutes.

[0038] In an embodiment, the method further comprises radioimaging the subject by PET, SPECT and/or CT, preferably after each treatment cycle.

[0039] In an embodiment, a positive PET, SPECT and/or CT scan is based on a visualisation of ^{64}Cu -SAR-bisPSMA (formula (I)) PET/SPECT/CT scan, where ^{64}Cu -SAR-bisPSMA uptake (standardized uptake value [SUV] max) of at least 1 known lesion is higher than that of the liver on the 1 hour PET/CT / SPECT scan.

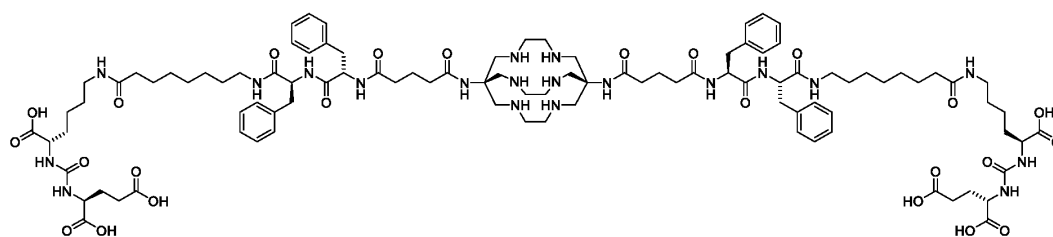
[0040] Additionally then, the present invention provides a method of radioimaging a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{64}Cu radioisotope:



Formula (I)

wherein the cancer is associated with expression or overexpression of a PSMA membrane protein, and wherein the dose of radiation delivered by the radioisotope is about 200 MBq/kg.

[0041] In certain embodiments of the above aspects, the compound of Formula (I) has the structure of Formula (Ia):



Formula (Ia)

[0042] In an embodiment of the above aspect, the method further comprises the step of obtaining images of the subject by PET/ SPECT and/or CT.

[0043] The present inventors believe that the images of the subject obtained by PET/SPECT and/or CT after administration of the ^{64}Cu -labelled compound of Formula (I) in accordance with the above aspects may dictate the corresponding dose of the ^{67}Cu -labelled compound of Formula (I) that is used for the treatment of the cancer associated with expression or overexpression of the PSMA membrane protein. Accordingly, one of the benefits of the present invention is that the same bis-compound may be used in a complete diagnosis-therapy regime by replacing the Cu radioisotope from ^{64}Cu to ^{67}Cu .

[0044] Thus the present inventors also believe that administration of more than one dose (i.e. multiple treatment cycles) of the aqueous formulations described herein for the treatment of a cancer associated with overexpression of the PSMA membrane antigen leads to greater accumulation of the radioisotope at the target site. Without wishing to be bound by theory, the present inventors believe that the use of the radiolabelled compounds described herein allow for greater doses of radiation to be delivered without an increase in the expected adverse effects. This therefore leads to greater efficacy in treatment. The diagnosis methods disclosed herein can be used before, or between treatment cycles in order to assess the effectiveness of the treatment.

[0045] Without wishing to be bound by theory, the present inventors believe that administration of more than one dose of the formulations described herein for the treatment of a cancer associated with overexpression of PSMA membrane protein leads to higher absorbed radiation doses at the cancer site, which leads to greater efficacy of treatment. This means that repeat administrations of the formulation containing the compound of Formula (I) complexed with a radioisotope may lead to greater survival of the subject, when compared with single

administrations of the formulations as disclosed herein. The method of the second aspect may comprise administration of multiple doses of the aqueous formulation of the complex containing the compound of Formula (I) and the radioisotope. In an embodiment, the method comprises the sequential administration of more than one dose of the aqueous formulation described in the second aspect. In some embodiments, the sequential doses of the aqueous formulation are administered between about 6 weeks and about 16 weeks apart. In an embodiment, the sequential doses of the aqueous formulation are administered about 6 weeks apart. In an embodiment, the total dose of radiation delivered to the bone marrow of the subject is less than about 2 Gy. In another embodiment, the total dose of radiation delivered to the kidneys of the subject is less than about 23 Gy. In another embodiment, the total dose of radiation delivered to the submandibular gland of the subject is less than about 24 Gy.

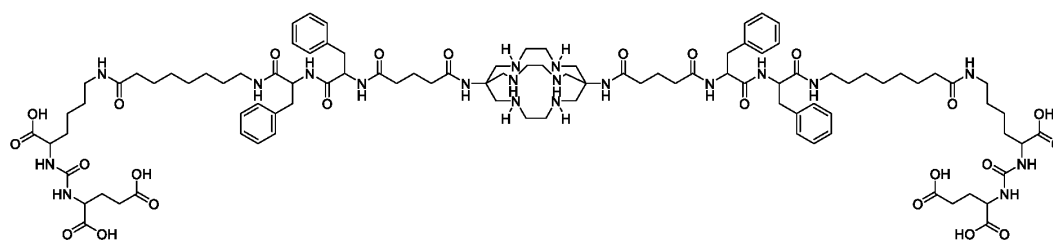
[0046] The methods disclosed herein may comprise administration of multiple doses of the complex containing the compound of Formula (I) and the radioisotope, where the doses administered are the same or different. In some embodiments, where multiple doses are administered, the second and any subsequent doses may be higher than the original dose. In some embodiments, multiple doses are administered where the doses are the same. In another embodiment, multiple doses are administered where the doses are different. One skilled in the art would understand that since methods discussed herein incorporate the use of a radioisotope, there is a maximum total dose of radiation that a subject may be given. In some embodiments, multiple doses are administered until the cumulative dose of radiation delivered to the kidneys of the subject reaches about 23 Gy. In some embodiments, multiple doses are administered, until the cumulative dose of radiation delivered to the submandibular gland of the subject reaches about 24 Gy.

[0047] In an embodiment, the aqueous formulation is administered intravenously. In another embodiment, the aqueous formulation is administered by slow infusion. In a preferred embodiment, the aqueous formulation is administered intravenously by slow infusion, for instance between about 30-60 minutes.

[0048] The methods disclosed herein include the administration of a radioisotope that emits ionising radiation. Since the kidneys are responsible for blood filtration, the kidneys of a subject to which the formulations comprising the compound of Formula (I) and a radioisotope has been administered are at risk of absorbing unwanted radiation as a result of active

reabsorption and retention of the radiolabelled compound of Formula (I). Prevention of nephrotoxicity may be achieved by co-administration of cationic amino acids that competitively inhibit the reabsorption of the compound of Formula (I) and thus the radioisotope. In some embodiments, the method of the second aspect further comprises the administration of a formulation containing one or more amino acids, or salts thereof. In some embodiments, one or more amino acids are in a cationic form. In some embodiments, the formulation containing one or more amino acids comprises lysine, or a salt thereof. In other embodiments, the formulation containing one or more amino acids comprises arginine, or a salt thereof. In a preferred embodiment, the method comprises the administration of a formulation comprising lysine and arginine, or salts thereof.

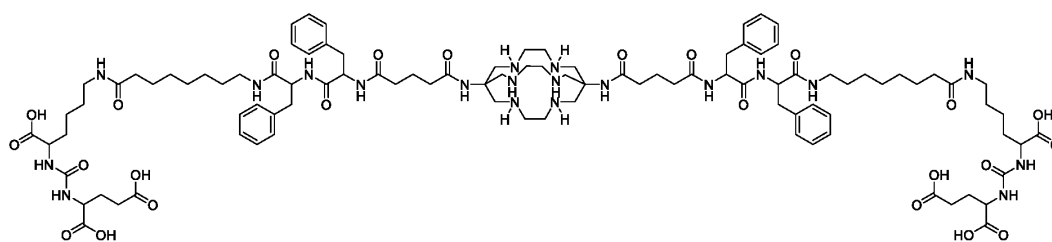
[0049] In a further aspect, the present invention provides a method for determining the dose of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope for use in the treatment of a cancer associated with expression or overexpression of the PSMA membrane protein in a subject in need thereof, the method comprising administering to the subject an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{64}Cu radioisotope:



Formula (I)

[0050] Without wishing to be bound by theory, the present inventors believe that the approach defined in the second aspect provides a more targeted method for the treatment of a cancer associated with expression or overexpression of the PSMA membrane protein in a subject. Any reference to the expression of the PSMA membrane protein as used herein also refers to the overexpression of the same protein.

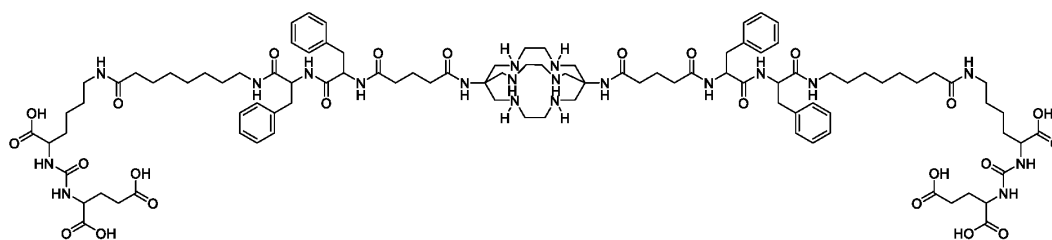
[0051] In another aspect, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of a PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer and/or mediate a decrease in prostate specific antigen (PSA) levels.

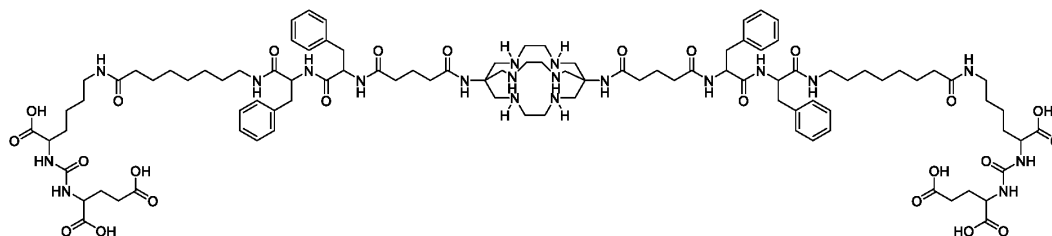
[0052] In another aspect, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of a PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer.

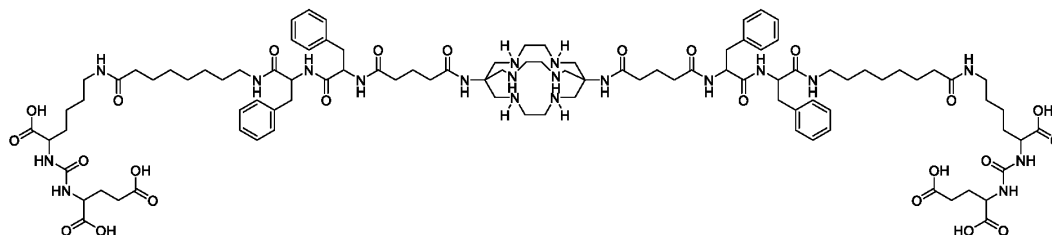
[0053] In another aspect, the present invention also provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for reducing PSA levels in a subject, wherein the PSA levels of the subject prior to administration of the aqueous formulation is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present.

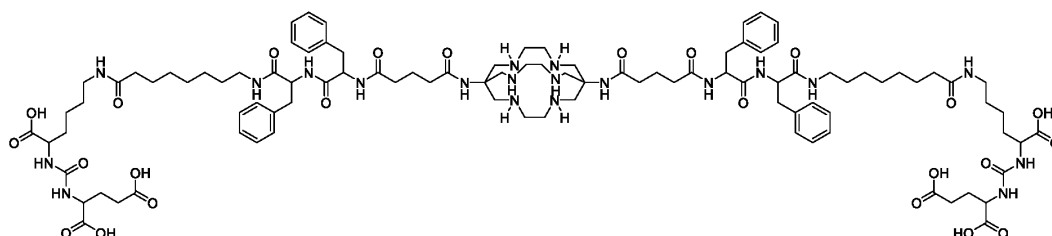
[0054] In another aspect, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for reducing PSA levels in a subject, wherein the PSA levels of the subject prior to administration of the aqueous formulation is greater than about 0 ng/ml, wherein the prostate gland of the subject is absent.

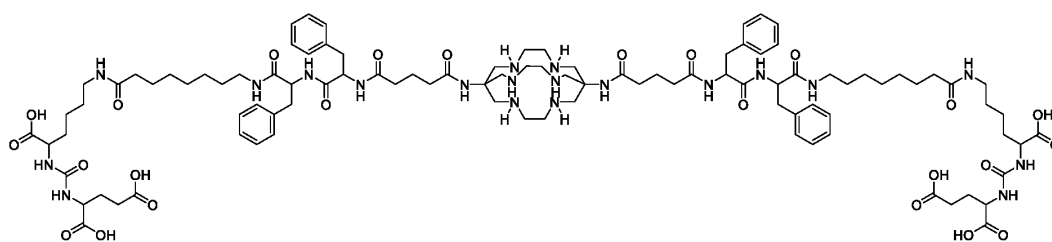
[0055] In a further embodiment, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of a PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 4 GBq.

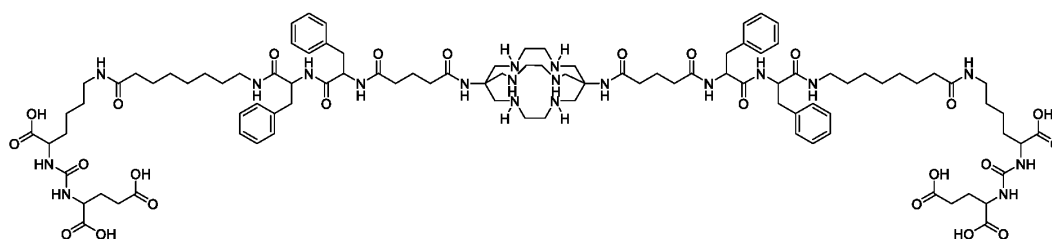
[0056] In a further embodiment, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of a PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 8 GBq.

[0057] In a further embodiment, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:

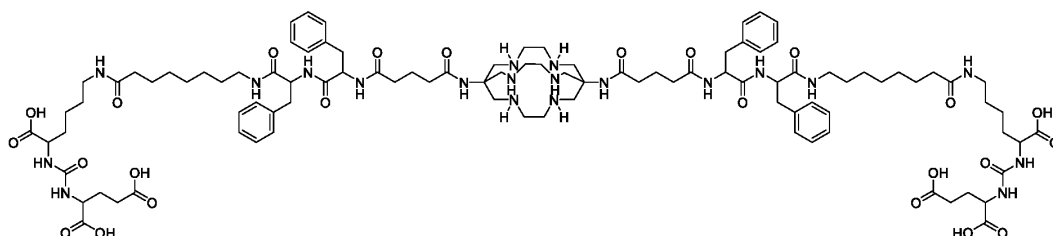


Formula (I)

in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of a PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 12 GBq.

[0058] In some embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope in the formulation is about 4 GBq, about 8 GBq, about 12 GBq, about 16 GBq, about 20 GBq or about 24 GBq. In some embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope in the formulation is more than about 12 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is more than about 16 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is more than about 20 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope is more than about 24 GBq. In other embodiments, the dose of radiation delivered by the ^{67}Cu radioisotope in the formulation is the highest dose that is tolerated by the subject.

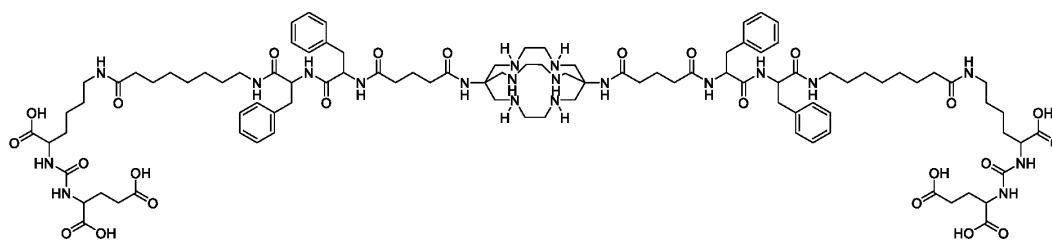
[0059] In a further aspect, the present invention provides use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for the treatment of a cancer associated with expression or overexpression of a PSMA membrane protein.

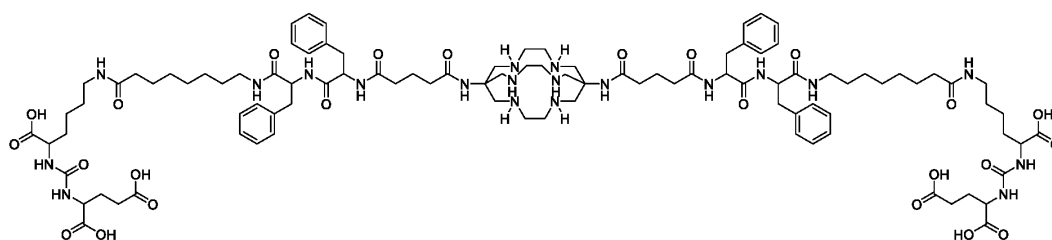
[0060] In another aspect, the present invention also provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for the treatment of a cancer associated with expression or overexpression of PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer.

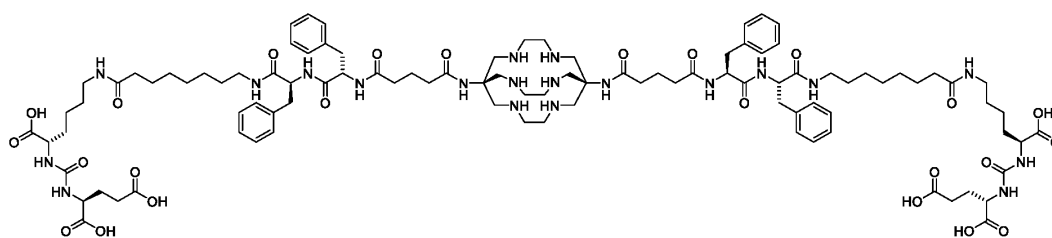
[0061] In a further aspect, the present invention also provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for the treatment of a cancer associated with expression or overexpression of PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer and/or mediate a decrease in prostate specific antigen (PSA) levels.

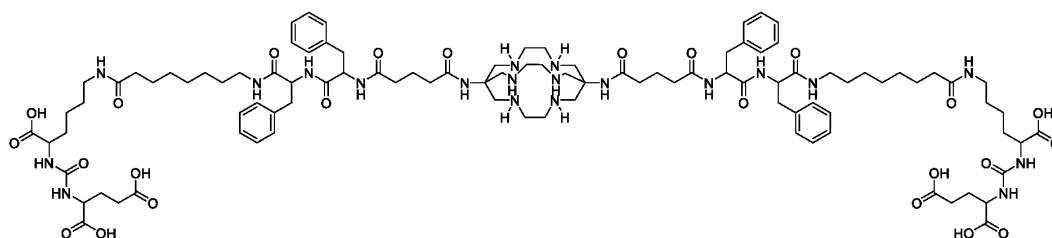
[0062] In certain embodiments of the above aspects, the compound of Formula (I) has the structure of Formula (Ia):



Formula (Ia)

[0063] In an embodiment of the above aspects, the cancer associated with expression or overexpression of a PSMA membrane protein is prostate cancer. In another embodiment, the cancer is metastatic prostate cancer. In a further embodiment, the prostate cancer is PSMA-expressing metastatic castrate resistant prostate cancer. In other embodiments, the prostate cancer shows resistance to prior androgen deprivation therapy and/or treatment with an androgen receptor pathway inhibitor. In some embodiments, the androgen receptor pathway inhibitor is enzalutamide or abiraterone.

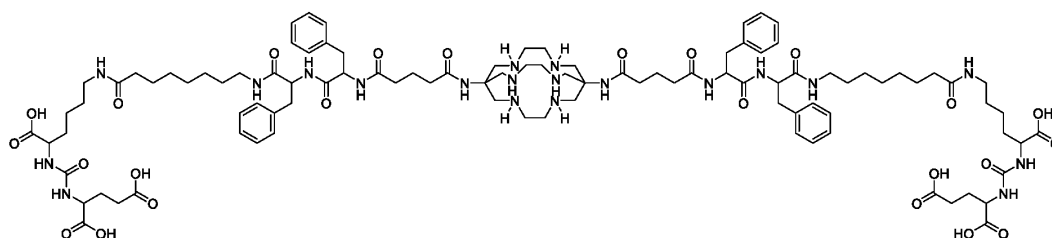
[0064] In accordance with another aspect, the present invention provides a the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for reducing prostate specific antigen (PSA) levels in a subject in need thereof, wherein the PSA levels of the subject prior to administration is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present.

[0065] In another aspect, the present invention provides the use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for reducing prostate specific antigen (PSA) levels in a subject in need thereof, wherein the PSA levels of the subject prior to administration is greater than about 0 ng/ml, wherein the prostate gland of the subject is absent.

Brief description of the figures

[0066] **Figure 1.** (A) PET-CT imaging of a subject having a T4 vertebral lesion after administration of ^{64}Cu -SARbisPSMA. (B) SPECT-CT imaging of the same subject having a T4 vertebral lesion 48 hours after administration of a first cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq. (C) SPECT-CT imaging of the same subject having a T4 vertebral lesion 48 hours after administration of a third cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq. (D) SPECT-CT imaging of the same subject having a T4 vertebral lesion 48 hours after administration of a fourth cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq.

[0067] **Figure 2.** (A) PET-CT imaging of a subject having a C1 vertebral lesion after administration of ^{64}Cu -SARbisPSMA. (B) SPECT-CT imaging of the same subject having a C1 vertebral lesion 48 hours after administration of a first cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq. (C) SPECT-CT imaging of the same subject having a C1 vertebral lesion 48 hours after administration of a third cycle of treatment with ^{67}Cu SAR-

bisPSMA at a dose of 4 GBq. (D) SPECT-CT imaging of the same subject having a C1 vertebral lesion 48 hours after administration of a fourth cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq.

[0068] Figure 3. (A) PET-CT imaging of a subject having a lesion in the right scapula after administration of ^{64}Cu -SARbisPSMA. (B) SPECT-CT imaging of the same subject having a lesion in the right scapula 48 hours after administration of a first cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq. (C) SPECT-CT imaging of the same subject having a lesion in the right scapula 48 hours after administration of a third cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq. (D) SPECT-CT imaging of the same subject having a lesion in the right scapula 48 hours after administration of a fourth cycle of treatment with ^{67}Cu SAR-bisPSMA at a dose of 4 GBq.

[0069] Figure 4. Graph showing the PSA levels (ng/ml) of a subject before treatment with ^{67}Cu -Sar-bisPSMA (dashed lines) and at various timepoints after a first and second administration of ^{67}Cu -Sar-bisPSMA. Administration of ^{67}Cu -Sar-bisPSMA resulted in a reduction in PSA level from 47.2 ng/ml (baseline, prior to administration) to 0.3 ng/ml. Administration of a second dose of ^{67}Cu -Sar-bisPSMA resulted in undetectable levels of PSA in the subject, where the lower detection limit is 0.05 ng/ml.

[0070] Figure 5. PET images showing the uptake of ^{64}Cu -Sar-bisPSMA in prostate cancer prior to treatment with ^{67}Cu -Sar-bisPSMA showing the presence of lesions (left, [SUV_{max}] 140.1) and after 2 administrations of ^{67}Cu -Sar-bisPSMA (right). Imaging prior to treatment clearly shows the presence of lesions in the subject, which are not visualised in the comparative image taken after treatment with 2 cycles of ^{67}Cu -Sar-bisPSMA (i.e. no uptake of ^{64}Cu -Sar-bisPSMA). This indicates that a complete response (i.e. the absence of detectable cancer after treatment) is observed after administration of ^{67}Cu -Sar-bisPSMA.

[0071] Figure 6. PET images of a patient with metastatic castrate resistant prostate cancer showing uptake of ^{64}Cu -Sar-bisPSMA before treatment (A) and after 2 cycles of treatment with ^{67}Cu -Sar-bisPSMA at 8 GBq per dose (B). The image taken prior to treatment shows the presence of a lesion (see arrow), while the image taken after treatment shows the reduction in size of the same lesion.

[0072] **Figure 7.** CT images of a patient with metastatic castrate resistant prostate cancer showing uptake of ^{64}Cu -Sar-bisPSMA before treatment (**A**) and after 2 cycles of treatment with ^{67}Cu -Sar-bisPSMA at 8 GBq per dose (**B**). The image taken prior to treatment clearly shows the presence of a lesion (see arrow), while the same lesion is not visualised under the same conditions after treatment with ^{67}Cu -Sar-bisPSMA.

[0073] **Figure 8.** PET/CT images of a patient with metastatic castrate resistant prostate cancer showing uptake of ^{64}Cu -Sar-bisPSMA before treatment (**A**) and after 2 cycles of treatment with ^{67}Cu -Sar-bisPSMA at 8 GBq per dose (**B**). The image taken prior to treatment clearly shows the presence of a lesion (see arrow), while the same lesion is not visualised under the same conditions after treatment with ^{67}Cu -Sar-bisPSMA.

Detailed description

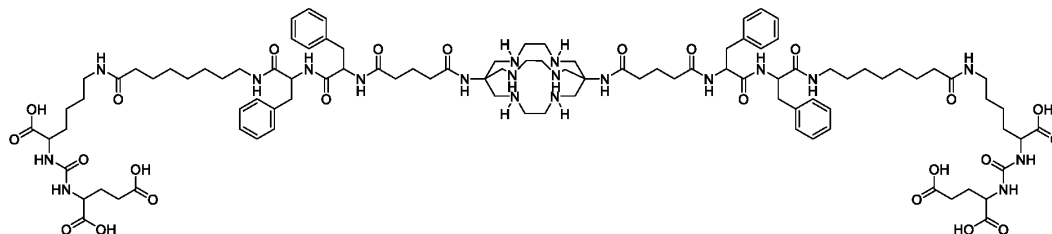
[0074] Throughout this specification and the claims which follow, unless the context requires otherwise, the word "comprise", and variations such as "comprises" and "comprising", will be understood to imply the inclusion of a stated integer or step or group of integers or steps but not the exclusion of any other integer or step or group of integers or steps.

[0075] The term "about" or "approximately" as used herein means within an acceptable error range for the particular value as determined by one of ordinary skill in the art, which will depend in part on how the value is measured or determined, i.e., the limitations of the measurement system.

[0076] Unless defined otherwise, all technical and scientific terms used herein have the same meaning as commonly understood by those of ordinary skill in the art to which the invention belongs. For the purposes of the present invention, the following terms are defined below.

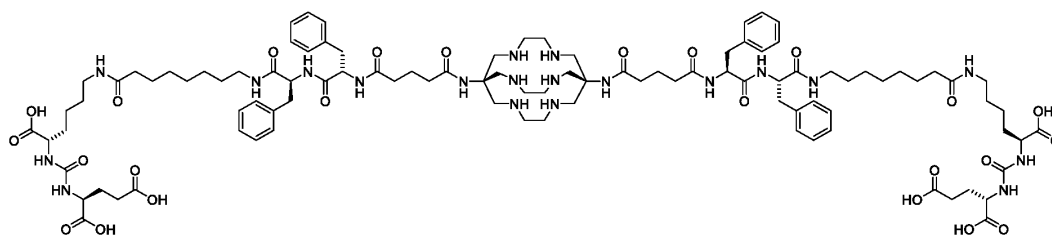
[0077] The compound of Formula (I) contains two ligands bearing a urea functional group and a sarcophagine, where the two ligands are bound to the sarcophagine by linker groups to provide a symmetrical molecule. The compound of Formula (I) is capable of binding to the PSMA membrane antigen, which is often overexpressed in cancerous tissue, specifically prostate tissue. The compound may be referred to as "Sar-bisPSMA" and contains a

macrocyclic sarcophagine fragment (i.e. 5-[[8-amino-3,6,10,13,16,19-hexaazabicyclo-[6.6.6]eico-1-yl)amino]-5-oxo-pentanyl) and two urea fragments (i.e. lysine-urea-serine). The compound of Formula (I) has the following structure:



Formula (I)

[0078] The compound of Formula (I) contains multiple stereocentres. All stereoisomers, for example enantiomers and diastereomers, of the compound and its salts are also contemplated by the present invention. In certain embodiments, the compound of Formula (I) has the structure of Formula (Ia):



Formula (Ia)

[0079] The term "pharmaceutically acceptable salts" refers to salts that retain the desired biological activity of the above-identified compounds, and include pharmaceutically acceptable acid addition salts and base addition salts. Suitable pharmaceutically acceptable acid addition salts of compounds of Formula (I) may be prepared from an inorganic acid or from an organic acid. Examples of such inorganic acids are hydrochloric acid, sulfuric acid, phosphoric acid, methane sulfonic acid, camphor sulfonic acid, oxalic acid, maleic acid, succinic acid, citric acid, formic acid, hydrobromic acid, benzoic acid, tartaric acid, fumaric acid, salicylic acid, mandelic acid, and carbonic acid. Appropriate organic acids may be selected from aliphatic, cycloaliphatic, aromatic, heterocyclic carboxylic and sulfonic classes of organic acids, examples of which are formic, acetic, propionic, succinic, glycolic, gluconic, lactic, malic, tartaric, citric, fumaric, maleic, alkyl sulfonic and arylsulfonic acids. Pharmaceutically acceptable salts also include those in which the main compound functions as

an acid and is reacted with an appropriate base to form, e.g., sodium, potassium, calcium, magnesium, ammonium, and choline salts. Those skilled in the art will further recognize that acid addition salts may be prepared by reaction of a compound with the appropriate inorganic or organic acid via any of a number of known methods. Alternatively, alkali and alkaline earth metal salts can be prepared by reacting a compound with the appropriate base via a variety of known methods. The following are further examples of acid salts that can be obtained by reaction with inorganic or organic acids: acetates, adipates, alginates, citrates, aspartates, benzoates, benzenesulfonates, bisulfates, butyrates, camphorates, digluconates, cyclopentanepropionates, dodecylsulfates, ethanesulfonates, glucoheptanoates, glycerophosphates, hemisulfates, heptanoates, hexanoates, fumarates, hydrobromides, hydroiodides, 2-hydroxy-ethanesulfonates, lactates, maleates, methanesulfonates, nicotines, 2-naphthalenesulfonates, oxalates, palmoates, pectinates, persulfates, 3-phenylpropionates, picrates, pivalates, propionates, succinates, tartrates, thiocyanates, tosylates, mesylates and undecanoates. Additional information on pharmaceutically acceptable salts can be found in Remington's Pharmaceutical Sciences, 19th Edition, Mack Publishing Co., Easton, PA 1995. In the case of agents that are solids, it is understood by those skilled in the art that the inventive compounds, agents and salts may exist in different crystalline or polymorphic forms, all of which are intended to be within the scope of the present invention and specified formulae.

[0080] The formulations of this invention for injection comprise pharmaceutically acceptable sterile aqueous solutions. Examples of suitable aqueous and nonaqueous carriers, diluents, solvents or vehicles include water, ethanol, polyols (such as glycerol, propylene glycol, polyethylene glycol, and the like), and suitable mixtures thereof, vegetable oils (such as olive oil), and injectable organic esters such as ethyl oleate. The formulations may also contain adjuvants such as preservative, wetting agents, emulsifying agents, and dispersing agents. Prevention of the action of micro-organisms may be ensured by the inclusion of various antibacterial and antifungal agents, for example, paraben, chlorobutanol, phenol sorbic acid, and the like. It may also be desirable to include isotonic agents such as sugars, sodium chloride, and the like. Prolonged absorption of the injectable pharmaceutical form may be brought about by the inclusion of agents that delay absorption such as aluminium monostearate and gelatin. The injectable formulations can be sterilized, for example, by filtration through a bacterial-retaining filter, or by incorporating sterilizing agents in the form of sterile solid compositions that can be dissolved or dispersed in sterile water or other sterile injectable medium just prior to use. The pharmaceutical formulation may further comprise a pH controller. Examples of

suitable pH controllers include hydrochloric acid, sodium hydroxide and the like. Identification of preferred pH ranges (where appropriate) and suitable excipients is routine in the art, for example, as described in Katdare and Chaubel (2006) *Excipient Development for Pharmaceutical, Biotechnology and Drug Delivery Systems* (CRC Press).

[0081] The formulations of the invention as disclosed herein may be provided in a pharmaceutically acceptable carrier or diluent. As will be appreciated by those skilled in the art, the choice of pharmaceutically acceptable carrier or diluent will be dependent on the route of administration and on the nature of the condition and subject to be treated. The particular carrier or diluent and route of administration may be readily determined by a person skilled in the art. The carrier or diluent and route of administration should be carefully selected so as to ensure activity of the compound of Formula (I) upon arrival at the site of action.

[0082] The pharmaceutical forms suitable for injectable use include sterile injectable solutions or dispersions and sterile powders for the preparation of sterile injectable solutions. Such forms should be stable under the conditions of manufacture and storage and may be preserved against reduction, oxidation and microbial contamination. For injection, compositions of the invention may be formulated in aqueous solutions, suitably in physiologically compatible buffers such as Hanks' solution, Ringer's solution or physiological saline buffer.

[0083] For the treatment of a cancer associated with expression or overexpression of the PSMA membrane protein, the compound of Formula (I) is complexed with a radioisotope. The present inventors have found that the sarcophagine fragment of Formula (I) has a strong affinity for copper isotopes and is capable of complexing and retaining a radioisotope for a time that is sufficient for the purposes of treatment, even after administration to a subject. The compound of Formula (I) may be complexed with a copper isotope. In an embodiment, the compound of Formula (I) is complexed with a ^{67}Cu radioisotope to provide a complex that can be used in the treatment of a cancer associated with expression or overexpression of the PSMA membrane protein. The half-life of the ^{67}Cu radioisotope is approximately 60 hours and undergoes beta decay, thus making the isotope suitable for localised radiotherapy. Since decay of the ^{67}Cu radioisotope is accompanied by gamma radiation, the treatment of a subject to which the compound of Formula (I) complexed with ^{67}Cu may be monitored and imaged by single-photon emission computed tomography (SPECT). In an embodiment, the method for treating a subject in need thereof by administration of a compound of Formula (I) complexed with ^{67}Cu includes

monitoring and/or imaging by SPECT. Other imaging techniques during treatment may also be used, for example, MRI and CT. In a preferred embodiment, the method for treatment includes imaging by SPECT and/or CT. Any reference to the expression of the PSMA membrane protein as used herein also refers to the overexpression of the same protein.

[0084] For the purposes of radioimaging, the dose of radiation delivered should be sufficient to provide images of sufficient quality without administering an excess amount to the patient. The dose of radiation (and subsequently the amount of the radiolabelled compound of Formula (I) complexed with a ^{64}Cu radioisotope) to be administered for the purposes of radioimaging may be determined based on the bodyweight of the subject. For the purposes of treatment, the dose of radiation to be administered and delivered to the subject by a ^{67}Cu radioisotope may be determined based on both bodyweight of the subject and the quality of the images obtained via radioimaging after administration of the compound of Formula (I) complexed with a ^{64}Cu radioisotope. In some embodiments, the radiolabelled compound of Formula (I) complexed with a ^{64}Cu radioisotope is used to model the distribution of the corresponding compound of Formula (I) complexed with a ^{67}Cu radioisotope. In some embodiments, radioimaging of a subject is performed after administration of a compound of Formula (I) complexed with a ^{64}Cu radioisotope. In other embodiments, radioimaging of a subject is performed after administration of a compound of Formula (I) complexed with a ^{67}Cu radioisotope. In some embodiments, radioimaging of a subject is performed after administration of a compound of Formula (I) complexed with a ^{67}Cu radioisotope, where the ^{67}Cu radiolabelled compound of Formula (I) is administered for the purposes of therapy.

[0085] As seen in Figures 1 to 3, the present inventors have found that the administration of a compound of Formula (I) radiolabelled with a ^{67}Cu radioisotope allows for a radioimaging to be performed up to 96 hours after the administration of the compound. Without wishing to be bound by theory, the present inventors believe that the compounds of Formula (I) as described herein show increased retention in a subject such that radioimaging of the subject is possible for a time after administration of the radiolabelled compound. In certain embodiments, radioimaging of a subject is performed after administration of a compound of Formula (I) radiolabelled with a ^{67}Cu radioisotope. In some embodiments, radioimaging of a subject is performed about 6 hours, about 12 hours, about 24 hours, about 36 hours, about 48 hours, about 60 hours, about 72 hours, about 84 hours or about 96 hours after administration of a compound of Formula (I) radiolabelled with a ^{67}Cu radioisotope.

[0086] The units of radioactivity as recited herein are given in gray (Gy) or becquerel (Bq). It will be appreciated that the dose of radiation may be converted from one unit to another using known conversion factors and that other units for the amount of radioactivity that are not explicitly recited herein may also be used.

[0087] As used herein the terms "treating", "treatment", "preventing", "prevention" and grammatical equivalents refer to any and all uses which remedy the stated cancer, prevent, retard or delay the establishment of the disease, or otherwise prevent, hinder, retard, or reverse the progression of the disease. Thus the terms "treating" and "preventing" and the like are to be considered in their broadest context. For example, treatment does not necessarily imply that a patient is treated until total recovery. Where the disease displays or is characterized by multiple symptoms, the treatment or prevention need not necessarily remedy, prevent, hinder, retard, or reverse all of said symptoms, but may prevent, hinder, retard, or reverse one or more of said symptoms.

[0088] As used herein, the term "cancer" broadly encompasses neoplastic diseases characterised by abnormal cell growth with the potential to invade or spread to other parts of the body. The cancer may be benign, which does not spread to other parts of the body. The cancer may be malignant, meaning that the cancer cells can spread through the circulatory system or lymphatic system. The term as used herein includes all malignant, i.e. cancerous, disease states. The cancer may be present as a tumour.

[0089] As used herein, the term "tumour" refers to any malignant cancerous or pre-cancerous cell growths. The term may also include leukemias, but is particularly directed to solid tumours or carcinomas. Where the cancer is present in the prostate, the cancer is termed "prostate cancer", which is typically characterised by increasing and/or elevated levels of serum prostate-specific antigen (PSA) and expression or overexpression of a PSMA membrane protein. A subject may have prostate cancer, where the cancer is a primary cancer and localised in the prostate gland. The prostate cancer may metastasise and spread to other parts of the subject. A subject may also have recurrent prostate cancer, which is characterised by an increase in PSA within 10 years of primary prostate cancer treatment.

[0090] In certain embodiments the method is effective in treating prostate cancer and in particular PSMA-expressing metastatic castrate resistant prostate cancer (mCRPC). In certain embodiments the mCRPC is progressive mCRPC despite prior androgen deprivation therapy and at least either enzalutamide and/or abiraterone (or other such androgen receptor pathway inhibitors).

[0091] Documented progressive mCRPC is based on at least one of the following criteria:

- i) serum/plasma prostate specific antigen (PSA) progression defined as 2 consecutive increases in PSA over a previous reference value measured at least 1 week prior, the minimal value being 0.1 ng/mL;
- ii) soft-tissue progression defined as a $\geq 20\%$ increase in the sum of the diameter (SOD) (short axis for nodal lesions and long axis for non-nodal lesions) of all target lesions based on the smallest SOD since the last treatment directed at the metastatic cancer has started (not including hormonal therapy) or the appearance of one or more new lesions; and
- iii) progression of bone disease: evaluable disease or new bone lesions(s) by bone scan.

[0092] The term "subject" as used herein refers to mammals and includes humans, primates, livestock animals (e.g. sheep, pigs, cattle, horses, donkeys), laboratory test animals (e.g. mice, rabbits, rats, guinea pigs), performance and show animals (e.g. horses, livestock, dogs, cats), companion animals (e.g. dogs, cats) and captive wild animals. Preferably, the mammal is human or a laboratory test animal. Even more preferably, the mammal is a male human.

[0093] The term "therapeutically effective amount" or "effective amount" is an amount sufficient to effect beneficial or desired clinical results. An effective amount can be administered in one or more administrations. For the purposes of radioimaging, an effective amount is sufficient for an image showing the localisation of the compound of Formula (I) administered to the subject, owing to the detection of the products of decay from the radioisotope that is complexed with the compound. For the purposes of treatment, an effective amount is typically sufficient to palliate, ameliorate, stabilize, reverse, slow and/or delay the progression of the cancer.

[0094] Radiological progression free survival (rPFS) is defined as the time from the date of the first treatment with ⁶⁷Cu-SAR-bisPSMA to radiographic progression on bone scan or radiographic soft tissue progression or death from any cause, whichever comes first. rPFS will be presented with a Kaplan-Meier curve and summary statistics (median and rPFS at 6, 9, and 12 months). In certain embodiments the method provides a rPFS of from over 6 months to over 5 years.

[0095] In some embodiments the effective amount is an amount that is effective to elicit one of the following:

Complete Response (CR): Disappearance of all target lesions. All pathological lymph nodes must have decreased to <10mm in short axis. Disappearance of all target lesions. Any pathological lymph nodes (whether target or non-target) must have reduction in short axis to <10 mm;

Partial Response (PR): At least a 30% decrease in the sum of diameters of target lesions, taking as reference the baseline sum diameters.

[0096] The treatment regime will typically involve a number of cycles (e.g. 1, 2, 3, 4, 5, or 6) of treatment with the cycles being continued until such time as the condition has been ameliorated. Once again the optimal number of cycles and the spacing between each treatment cycle will depend upon a number of factors such as the height and weight of the subject, the severity of the condition being treated, the health (or lack thereof) of the subject being treated and their previous reactions to radiotherapy and/or the extent of the condition as determined through radioimaging.

[0097] The present inventors have found that a treatment regime having one or more cycles of treatment with a radiolabelled compound of Formula (I) results in the treatment of a subject as shown by a reduced uptake of the compound as visualised in Figures 1 to 3. Since reduced uptake of the compound indicates a lower concentration or expression of the PSMA receptor, the present inventors believe that the administration of a radiolabelled compound of Formula (I) in accordance with one or more methods as disclosed herein results in the treatment of a prostate cancer in a subject in need thereof.

[0098] The formulations defined in the present specification for methods of treatment may be administered parenterally, with intravenous administration preferred. In an embodiment, the

aqueous formulation comprising a radiolabelled compound of Formula (I) is administered intravenously, either by bolus administration or slow infusion. In another embodiment, the compound of Formula (I) radiolabelled with a ^{67}Cu radioisotope is administered intravenously by slow infusion.

[0099] It will be understood that the specific dose of the radiolabelled compound of Formula (I) for any particular subject will depend upon a variety of factors including, for example, the age, body weight and indication of the individual to be treated, the time of administration, rate of excretion, and combination with any other treatment or therapy. Single or multiple administrations can be carried out with dose levels and pattern being selected by the treating physician. A broad range of doses may be applicable. Dosage regimens may be adjusted to provide the optimum therapeutic response. For example, a given dose delivering a certain amount of radiation may be calculated as a fraction of the total radiation to be delivered to the subject. Dosage regimens may comprise the administration of multiple doses of the radiolabelled compound of Formula (I), where the doses are the same or different. In some embodiments, the methods for treatment of a cancer associated with overexpression of the PSMA membrane protein comprise the administration of multiple doses of a compound of Formula (I) complexed with a copper radioisotope, where the doses are the same. In other embodiments, the methods comprise the administration of multiple doses, where the second and subsequent doses are higher than the first dose administered to the subject. In some embodiments, the dose administered for the purpose of treatment or therapy is determined by prior radioimaging of the subject by administration of the compound of Formula (I) complexed with a ^{64}Cu radioisotope in order to locate the cancer sites, estimate the amount of the compound retained by the subject (and subsequently the amount of radioactivity delivered) and assess the nature of the cancer sites. The present inventors believe that the use of the compound of Formula (I) or a pharmaceutically acceptable salt thereof for both radioimaging and radiotherapy represents a theranostic approach to cancers associated with overexpression of the PSMA membrane protein. Without wishing to be bound by theory, the present inventors believe that the use of such a theranostic compound presents a more tailored approach to cancer therapy.

[0100] In combination with the methods for radioimaging a cancer associated with overexpression of a PSMA membrane protein where the compound of Formula (I) having a different isotope is administered, the methods disclosed herein represent a theranostic

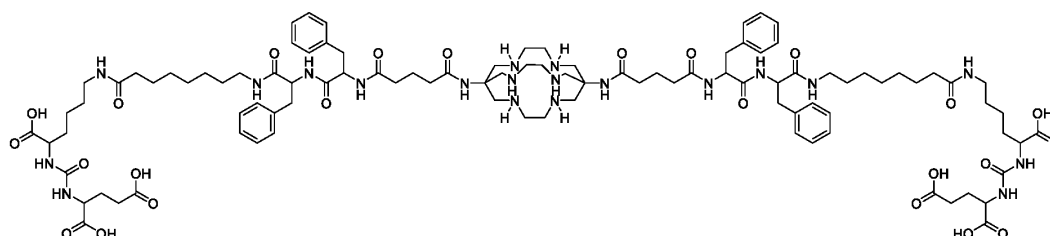
approach, i.e. a therapeutic and diagnostic approach, to the treatment of such cancers. This is because administration of the compound of Formula (I) may also be complexed with a radioisotope that allows for the radioimaging of a subject, while administration of the compound of Formula (I) that is complexed with ^{67}Cu allows for treatment of the subject. Radioimaging allows for visualisation of where the compound of Formula (I) accumulates, which then corresponds to the site of treatment. Without wishing to be bound by theory, the present inventors believe that the methods and uses disclosed herein allow for more efficacious treatment of cancers associated with overexpression of the PSMA membrane protein. The use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof in combination with a copper radioisotope allows for a higher dose of radioactivity to be delivered in a single dose. Since the compound of Formula (I) is specific for the PSMA membrane protein and retains the copper radioisotope for a longer time (when compared to other metal chelators), the radioactivity is delivered to the cancer site and localised more efficiently. The compound of Formula (I) also shows better clearance from key organs. This in turn reduces any off-target effects of the radioisotope and limits unwanted damage to healthy tissue attributed to dissociation and subsequent circulation of the radioisotope. Better clearance of the radiolabelled compound of Formula (I) and retention at the targeted cancer site leads to images of higher contrast and subsequently more reliable diagnostic images. The ability to deliver a more sustained radiation dose by administration of the compound of Formula (I) complexed with a copper radioisotope also leads to more efficient treatment overall, as smaller amounts of the compound of Formula (I) and the radioisotope are required. Where the requisite radiation is delivered in fewer doses, this results in better tolerance to treatment by the subject.

[0101] The methods of the present invention relate to the treatment of a cancer associated with overexpression of the PSMA membrane protein in a subject in need thereof. In an embodiment, the cancer is prostate cancer. In an embodiment, the cancer is non-metastatic prostate cancer. In an embodiment, the cancer is metastatic prostate cancer. In another embodiment, the prostate cancer is PSMA-expressing castrate resistant prostate cancer. In some subjects, the prostate cancer shows resistance to prior androgen deprivation therapy and/or prior treatment with an androgen receptor pathway inhibitor. Examples of such inhibitors include abiraterone, enzalutamide, apalutamide, and darolutamide. In some embodiments, the subject may display progression of PSA, i.e. an increase in the level of serum PSA. In some embodiments, the subject may display soft-tissue progression, which may manifest as an increase in the number of lesions detected and visualised by radioimaging according a method as defined herein. In

some embodiments, the subject may display progression of the cancer that manifests as progress of bone disease, for example, one or more bone lesions detected by bone scan. In some embodiments, the subject has received chemotherapy prior to treatment in accordance with one or more embodiments of the present invention. In other embodiments, the subject has received one or more lines of treatment (i.e. other treatment options) prior to treatment in accordance with one or more embodiments of the present invention. In some embodiments, the subject has received two, three, four or five lines of treatment prior to treatment in accordance with one or more embodiments of the present invention.

[0102] Prostate cancer is present in some subjects as metastatic castration resistant prostate cancer (mCPC), which is defined as a subject having prostate cancer that has spread or metastasized to other locations of the subject. While a decrease in PSA levels are initially observed after prostatectomy (i.e. removal of the prostate) of the subject and subsequent chemotherapy and/or radiotherapy, an increase in PSA levels is observed in some subjects as a result of one or more metastases. The present inventors have found that the treatment of subjects having mCPC characterised by rising PSA levels with a radiolabelled compound of Formula (I) results in a decrease in the PSA level of the subject. In conjunction with the results now presented in Figures 1 to 3 showing a reduction in the uptake of a radiolabelled compound of Formula (I) indicating a lower expression of PSMA associated with a cancer or tumor site, the present inventors believe that the administration of a radiolabelled compound of Formula (I) results in the treatment of a prostate cancer in a subject.

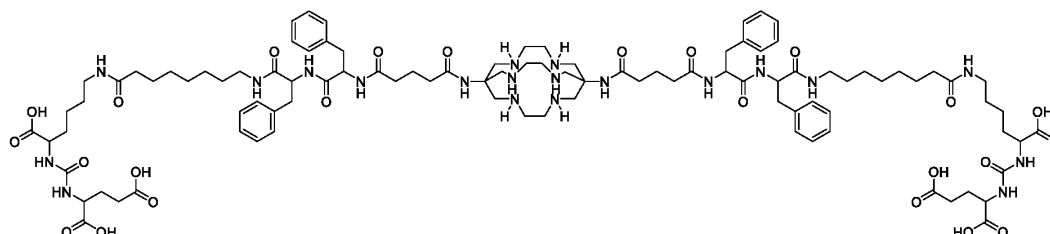
[0103] The present invention therefore provides a method for reducing the prostate specific antigen (PSA) level in a subject, the method comprising administering to the subject in need thereof a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount:



Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present..

[0104] The present invention also provides a method reducing the prostate specific antigen (PSA) level in a subject, the method comprising administering to the subject in need thereof a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount:



Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0.05 ng/ml, wherein the prostate gland of the subject is absent.

[0105] Without wishing to be bound by theory, the present inventors believe that the PSA levels in a subject prior to administration of the compound of Formula (I) complexed with a ^{67}Cu radioisotope will depend on whether the prostate is present or absent in the subject. For example, where a patient previously had the prostate removed, the detection of PSA in the blood of the patient likely indicates the presence of a prostate (or prostate-related) cancer. In other patients where the prostate is intact (i.e. present), the amount of PSA that is detected is relevant in whether a prostate (or prostate-related) cancer is present.

[0106] The PSA level of a subject is determined by one or more known analysis techniques, such as blood sampling and subsequent analysis. An increase or decrease in the PSA level of the subject is determined with respect to an initial level as determined prior to treatment with a radiolabelled compound of Formula (I). In some embodiments, the PSA level of the subject decreases after treatment with one or more doses of a radiolabelled compound of Formula (I), when compared to the PSA level of the subject prior to treatment. In certain embodiments, the PSA level of the subject decreases after one cycle of treatment with a radiolabelled compound of Formula (I). In certain embodiments, the PSA level of the subject decreases after two cycles of treatment with a radiolabelled compound of Formula (I). In certain embodiments, the PSA level of the subject decreases after three cycles of treatment with a radiolabelled compound of

Formula (I). In certain embodiments, the PSA level of the subject decreases after four or more cycles of treatment with a radiolabelled compound of Formula (I).

[0107] In certain embodiments, the PSA level of a subject decreases after one or more administrations of a radiolabelled compound of Formula (I). In certain embodiments, the PSA level of a subject decreases by about 10%, about 20%, about 30%, about 40%, about 50%, about 60%, about 70%, about 80%, about 90%, about 95% or about 99% relative to the PSA level of the same subject prior to treatment with a radiolabelled compound of Formula (I). In certain embodiments, the decrease in the PSA level of the subject is associated with reduced uptake of the radiolabelled compound of Formula (I) as determined by imaging with SPECT-CT or PET-CT.

[0108] In certain embodiments, the PSA level of the subject prior to administration is greater than about 0.1 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 1 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 10 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 100 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 500 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 1000 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 1500 ng/ml. In other embodiments, the PSA level of the subject prior to administration is greater than about 2000 ng/ml.

[0109] In some embodiments, the subject is administered more than one dose of the compound of Formula (I) complexed with a ^{67}Cu radioisotope. In certain embodiments, the subject is administered two doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope. In other embodiments, the subject is administered three doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope.

[0110] Where two doses of the compound of Formula (I) complexed with a ^{67}Cu radioisotope is administered, the second dose is administered at least four weeks after the first dose. In other embodiments, the second dose is administered at least six weeks after the first dose. In other embodiments, the second dose is administered at least 8 weeks after the first dose. In other embodiments, the second dose is administered at least 12 weeks after the first dose. In other

embodiments, the second dose is administered at least 6 months after the first dose. In other embodiments, the second dose is administered at least 12 months after the first dose. In other embodiments, the second dose is administered at least 18 months after the first dose. In other embodiments, the second dose is administered at least 24 months after the first dose. In other embodiments, the second dose is administered at least 30 months after the first dose. In other embodiments, the second dose is administered at least 36 months after the first dose. In other embodiments, the second dose is administered at least 4 years after the first dose. In other embodiments, the second dose is administered at least 5 years after the first dose.

[0111] In some embodiments, the PSA level of a subject decreases after two administrations of a radiolabelled compound of Formula (I) at a dose of 4 GBq each. In some embodiments, the PSA level of a subject decreases after three administrations of a radiolabelled compound of Formula (I) at a dose of 4 GBq each. In some embodiments, the PSA level of a subject decreases after four administrations of a radiolabelled compound of Formula (I) at a dose of 4 GBq each. In some embodiments, the PSA level of a subject decreases after two administrations of a radiolabelled compound of Formula (I) at a dose of 8 GBq each. In some embodiments, the PSA level of a subject decreases after three administrations of a radiolabelled compound of Formula (I) at a dose of 8 GBq each. In some embodiments, the PSA level of a subject decreases after four administrations of a radiolabelled compound of Formula (I) at a dose of 8 GBq each. In some embodiments, the PSA level of a subject decreases after two administrations of a radiolabelled compound of Formula (I) at a dose of 12 GBq each. In some embodiments, the PSA level of a subject decreases after three administrations of a radiolabelled compound of Formula (I) at a dose of 12 GBq each. In some embodiments, the PSA level of a subject decreases after four administrations of a radiolabelled compound of Formula (I) at a dose of 12 GBq each.

[0112] The present invention also contemplates combination therapies, wherein the radiolabelled compound of Formula (I) as described herein is co-administered with other suitable agents that may facilitate the desired therapeutic outcome. The term “co-administered” means simultaneous administration in the same formulation or in two different formulations via the same or different routes or sequential administration by the same or different routes. The term “concomitant” refers to the administration of more than one formulation, where the formulations are administered to the subject at the same time. The term “simultaneously” means that the active agents are administered at substantially the same time. The term

“sequential” administration means a time difference of from seconds, minutes, hours or days between administrations of the agents. Administration may be in any order.

[0113] Since the methods disclosed herein relate to the administration of a radioisotope that emits ionising radiation, co-administration of one or more amino acids with the aqueous formulations comprising a radiolabelled compound of Formula (I) as disclosed herein may prevent or limit nephrotoxicity resulting from retention of the radiopharmaceutical. The one or more amino acids co-administered to a subject undergoing treatment for a cancer associated with overexpression of a PSMA membrane protein competitively inhibit the reabsorption of the radiolabelled compound of Formula (I) by the proximal tubules of the kidneys. The present inventors believe that limiting reuptake of the radiolabelled compound of Formula (I) and therefore reducing nephrotoxicity of the subject allows for higher doses of the compound to be administered and therefore increase the efficiency of treatment. The methods for the treatment of a cancer as disclosed herein further comprise the administration of one or more amino acids, or salts thereof, to the subject. In one embodiment, a formulation comprising one or more amino acids or salts thereof is co-administered with an aqueous formulation comprising a compound of Formula (I) complexed with a ^{67}Cu radioisotope. In one embodiment, the one or more amino acids includes lysine or a salt thereof. In another embodiment, the one or more amino acids includes arginine or a salt thereof. In a preferred embodiment, the method for the treatment of a cancer further comprises the administration of lysine and/or arginine, or salts thereof. In a preferred embodiment, the method for the treatment of a cancer further comprises the administration of lysine and arginine or salts thereof. In a preferred embodiment, the method for the treatment of a cancer as disclosed herein further comprises the co-administration of lysine and arginine or salts thereof with the compound of Formula (I) complexed with a ^{67}Cu radioisotope. In some embodiments, the one or more amino acids, or salts thereof, are administered as an intravenous infusion. In some embodiments, the formulation comprising one or more amino acids comprises L-lysine or a salt thereof. In other embodiments, the formulation comprising one or more amino acids comprises L-arginine or a salt thereof. In some embodiments, the one or more amino acids are present as a hydrochloride salt. In some embodiments, the one or more amino acids are each present at a concentration of about 2.5% w/v.

[0114] In an embodiment, the method for the treatment of a cancer associated with overexpression of the PSMA membrane protein comprising administering to a subject in need

thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to with ^{67}Cu in a therapeutically effective amount, and an aqueous formulation of one or more amino acids. In a preferred embodiment, the formulations comprising the radiolabelled compound of Formula (I) complexed with ^{67}Cu and one or more amino acids are administered concurrently. In another preferred embodiment, the formulation comprising one or more amino acids is administered to the subject prior to the administration of the compound of Formula (I) complexed with ^{67}Cu .

[0115] As used herein, the term “amino acid” refers to a molecule which contains both an amino and a carboxyl functional group. The amino acid may be a natural or unnatural amino acid and may also be in equilibrium with its zwitterionic form. The amino acid may contain modifications at either the amino and/or carboxyl terminus, or may contain a free amino group or carboxyl group. Further modification of the amino acid side chain or additional substitutions at other parts of the amino acid are also contemplated.

[0116] As used herein, naturally occurring amino acids are the L- or D-form of the twenty amino acids commonly found in nature. These are glycine (Gly, G), alanine (Ala, A), valine (Val, V), leucine (Leu, L), isoleucine (Ile, I), methionine (Met, M), proline (Pro, P), phenylalanine (Phe, F), tryptophan (Trp, W), serine (Ser, S), threonine (Thr, T), asparagine (Asn, N), glutamine (Gln, Q), tyrosine (Tyr, Y), cysteine (Cys, C), lysine (Lys, K), arginine (Arg, R), histidine (His, H), aspartic acid (Asp, D), and glutamic acid (Glu, E).

[0117] The methods of the present invention may further comprise the administration of an anti-emetic agent. In one embodiment, the methods of the present invention further comprises the administration of an anti-emetic agent to the subject. In some embodiments, the anti-emetic is administered concurrently with or prior to the compound of Formula (I) complexed with ^{67}Cu .

[0118] The methods of treatment as disclosed herein comprise the administration of a formulation comprising a compound of Formula (I) complexed with ^{67}Cu . The formulation may be administered intravenously, for example, by slow intravenous infusion. The methods of treatment as disclosed herein may comprise a single administration of the formulation comprising a compound of Formula (I) complexed with ^{67}Cu , or more than one administration of the same or different formulation. In an embodiment, the method for the treatment of a

cancer comprises the administration of one dose of an aqueous formulation comprising a compound of Formula (I) complexed with ^{67}Cu . In another embodiment, the method comprises the administration of two doses of an aqueous formulation comprising a compound of Formula (I) complexed with ^{67}Cu . In another embodiment, the method comprises the administration of three doses of an aqueous formulation comprising a compound of Formula (I) complexed with ^{67}Cu . In another embodiment, the method comprises the administration of four doses of an aqueous formulation comprising a compound of Formula (I) complexed with ^{67}Cu .

[0119] Where more than one administration is required, the interval between doses of the formulation may be between about 1 week to about 14 weeks, or more. In an embodiment, the method comprises the administration of more than one dose of an aqueous formulation comprising a compound of Formula (I) complexed with ^{67}Cu , where the doses are administered about 1 week apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 2 weeks apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 4 weeks apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 6 weeks apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 8 weeks apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 10 weeks apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 12 weeks apart. In another embodiment, the method comprises the administration of more than one dose, where the doses are administered about 14 weeks apart. In some embodiments, the methods for treatment discussed herein comprise the administration of two or more doses of a formulation comprising a compound of Formula (I) complexed with ^{67}Cu , where the time between the doses may be the same. In some embodiments, the method comprises the administration of two or more doses of the formulation, where the time between doses is about the same, for example, about 1 week between each dose, about 2 weeks between each dose, about 4 weeks between each dose, about 6 weeks between each dose, about 8 weeks between each dose, about 10 weeks between each dose, about 12 weeks between each dose or about 14 weeks between each dose. In other embodiments, the time between doses may be different, for example, about 6 weeks between the first and second doses, and about 8 weeks between the second and third doses. Other embodiments where the time between different doses are also contemplated, where the time

between two sequential doses may be about 1 week, 2 weeks, 4 weeks, about 6 weeks, about 8 weeks, about 10 weeks, about 12 weeks, about 14 weeks or about 16 weeks.

[0120] In other embodiments, a second dose is administered at least four weeks after the first dose. In certain embodiments, a second dose is administered about six weeks after the first dose. In other embodiments, a second dose is administered about eight weeks after the first dose. In other embodiments, a second dose is administered about 12 weeks after the first dose. In some embodiments, a second dose is administered about 6 months, about 12 months, about 18 months, about 24 months, about 30 months or about 36 months after the first dose. In other embodiments, a second dose is administered about 4 years after the first dose. In other embodiments, a second dose is administered about 5 years after the first dose.

[0121] In an embodiment, the method for the treatment of a cancer associated with overexpression of the PSMA membrane protein comprises the administration of one dose of a formulation comprising a compound of Formula (I) complexed with ^{67}Cu . In some embodiments where the formulation comprising the compound of Formula (I) complexed with ^{67}Cu is administered more than once, the formulation that is administered on each occasion is the same or different. Where the formulation is different, the compound of Formula (I) complexed with ^{67}Cu in each formulation may deliver different doses of radioactivity, for example, about 4 GBq, about 8 GBq, about 12 GBq, about 16 GBq, about 20 GBq or about 24 GBq. In an embodiment, the aqueous formulation of a compound of Formula (I) complexed with ^{67}Cu that is administered for a method for the treatment of a cancer associated with overexpression of a PSMA membrane antigen delivers a dose of radiation between about 4 GBq to about 24 GBq to the subject. In an embodiment, the aqueous formulation delivers a dose of about 4 GBq. In another embodiment, the aqueous formulation delivers a dose of about 6 GBq. In another embodiment, the aqueous formulation delivers a dose of about 8 GBq. In another embodiment, the aqueous formulation delivers a dose of about 10 GBq. In yet another embodiment, the aqueous formulation delivers a dose of about 12 GBq. In a further embodiment, the aqueous formulation delivers a dose of about 16 GBq. In another embodiment, the aqueous formulation delivers a dose of about 20 GBq. In a further embodiment, the aqueous formulation delivers a dose of about 24 GBq.

[0122] In certain embodiments, the method for the treatment of a cancer associated with overexpression of the PSMA membrane protein comprises the administration of more than one

dose of the compound of Formula (I) complexed with a ^{67}Cu radioisotope, where each dose is about 4 GBq each. In other embodiments, each dose is about 8 GBq each. In other embodiments, each dose is about 12 GBq.

[0123] Alternatively, the dose of radioactivity that is delivered by the compound of Formula (I) complexed with ^{67}Cu is the maximum dose that is tolerated by the individual subject. One skilled in the art would understand that the maximum tolerated dose will vary among subjects. The present inventors have found that the administration of a compound of Formula (I) with a radioisotope that is suitable for imaging may allow not only for the distribution and uptake of the radiolabelled compound to be visualised, but also the tolerance the subject has for the given dose. Where the subject appears to tolerate the dose well and other physiological measures (e.g. liver and kidney function) are satisfactory, this information may be used to determine a higher dose of radiation that is specific to the subject. In an embodiment, the methods disclosed herein include the step of evaluating the tolerance of a subject to a dose of the compound of Formula (I) complexed with ^{67}Cu and modifying the dose of radioactivity that is delivered to the subject in a subsequent dose. Those skilled in the art will understand that a variety of techniques, including nuclear imaging, comparison between baseline levels of radiation and levels after administration of the compound of Formula (I) complexed with ^{67}Cu to determine uptake, comparison between the size and number of lesions before and after administration and monitoring of biochemical markers through one or more diagnostic assays of tissues may be used.

[0124] The present invention therefore also provides a method for the treatment of a cancer associated with overexpression of a PSMA membrane protein, where the dose of the compound of Formula (I) complexed with ^{67}Cu that is administered to the subject is specific to the subject and determined through a combination of imaging and physiological assay techniques. In an embodiment, the dose is determined by the administration of a compound of Formula (I) complexed with a suitable radioisotope to the subject and followed by imaging of the subject for a time to obtain one or more images that can be used to determine the suitability of the administered dose for the subject. In some embodiments, the imaging may be performed by one or more techniques, such as PET, SPECT and CT. Without wishing to be bound by theory, the present inventors believe that the methods disclosed herein allow for more sophisticated and personalised regimes for the treatment of a cancer in a subject that is associated with overexpression of a PSMA membrane protein.

[0125] The reference in this specification to any prior publication (or information derived from it), or to any matter which is known, is not, and should not be taken as an acknowledgment or admission or any form of suggestion that that prior publication (or information derived from it) or known matter forms part of the common general knowledge in the field of endeavour to which this specification relates.

[0126] Those skilled in the art will appreciate that the invention described herein 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 which fall within the spirit and scope. 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 of any two or more of said steps or features.

Examples

[0127] The following examples are illustrative of the disclosure and should not be construed as limiting in any way the general nature of the disclosure of the description throughout this specification.

Example 1 - ^{64}Cu -SAR-bisPSMA PET/CT scans

[0128] Quantitative PET/CT imaging is performed after each administration of ^{64}Cu -SAR-bisPSMA at the designated timepoints to confirm eligibility to receive therapy with ^{67}Cu -SAR-bisPSMA; to assess the biodistribution of ^{64}Cu -SAR-bisPSMA; or to perform dosimetry calculations. The ^{64}Cu -SAR-bisPSMA PET/CT scans may also be used to explore whether ^{64}Cu -SAR-bisPSMA is predictive of ^{67}Cu -SAR-bisPSMA dosimetry, biodistribution and potential therapeutic efficacy.

Example 2 - ^{67}Cu -SAR-bisPSMA SPECT/CT scans

[0129] Quantitative SPECT/CT imaging of ^{67}Cu -SAR-bisPSMA is used to assess the biodistribution of ^{67}Cu -SAR-bisPSMA or to perform dosimetry calculations. Quantitation is achieved by standard means.

Example 3 - Dosimetry of ^{64}Cu -SAR-bisPSMA and ^{67}Cu -SAR-bisPSMA

[0130] The radiation dosimetry may be determined (where necessary) using methods for radiation dose.

[0131] Dosimetry analysis utilizes the ^{64}Cu -SAR-bisPSMA PET/CT scans acquired at 1-, 4-, 12-, 24- and 48 hours post injection of ^{64}Cu -SAR-bisPSMA (additional later scans may be performed if adequate residual activity levels remain in the participant) to determine:

- i) Absorbed dose (mGy/MBq) in organs and effective dose (mSv/MBq) from ^{64}Cu -SAR-bisPSMA.
- ii) Modeled absorbed dose (mGy/MBq) in organs from ^{67}Cu -SAR-bisPSMA.
- iii) Modeled estimated total cumulative administered activity of ^{67}Cu -SAR-bisPSMA (GBq) that does not exceed the specific organ tolerance limits (23 Gy to the kidney, 2 Gy to the bone marrow and 24 Gy to the submandibular gland).

[0132] Dosimetry analysis from ^{67}Cu -SAR-bisPSMA SPECT/CT scans acquired at 1-, 4-, 12-, 24- and 48 hours following each administration of ^{67}Cu -SAR-bisPSMA (note: additional later scans may be performed if adequate residual activity levels remain in the participant) to determine the absorbed dose (mGy/MBq) in organs from ^{67}Cu -SAR-bisPSMA.

[0133] Copper-specific methodology was used to calculate the dosimetry for both ^{64}Cu -SAR-bisPSMA and ^{67}Cu -SAR-bisPSMA and the modelling of ^{67}Cu -SAR-bisPSMA dosimetry utilizing the ^{64}Cu -SAR-bisPSMA PET/CT data.

Example 4 – Determination of PSA levels

[0134] Blood tests are used to determine PSA levels of a patient, with up to 15 mL of blood obtained from a patient for analysis according to established protocols for determining the level of PSA in blood.

[0135] The PSA level of a subject is monitored by the same process at a later time, e.g. every 2 weeks, every 4 weeks, every 6 weeks, or the like. A further sample of blood is taken and analysed in the same manner. Comparison of the PSA level of the subject over time can be made.

Example 5 – Treatment with ^{67}Cu -SAR-bisPSMA – single dose

[0136] Generally, patients received an intravenous infusion of ^{67}Cu -Sar-bisPSMA over about 30 minutes, where the amount (i.e. volume) of the infusion is determined by the dose to be administered to the patient.

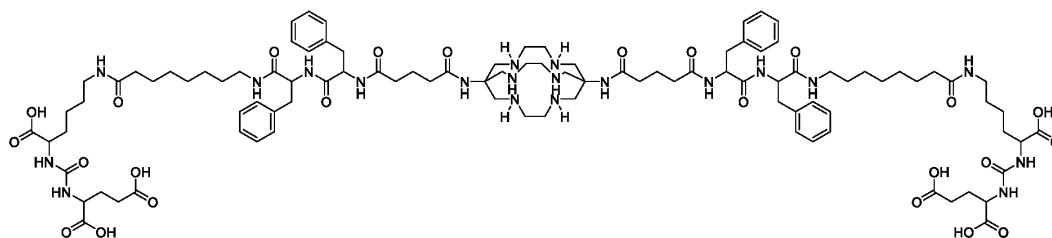
[0137] Six patients received a single administration of 4 GBq of ^{67}Cu -Sar-bisPSMA, with none of the patients reporting any dose-limiting toxicities (DLTs). Three patients received a single dose of 8 GBq of ^{67}Cu -Sar-bisPSMA, with none of the patients reporting any DLTs. Six patients received a single administration of 12 GBq of ^{67}Cu -Sar-bisPSMA, with none of the patients reporting any DLTs.

Example 6 – Treatment with ^{67}Cu -Sar-bisPSMA – multiple doses

[0138] Generally, patients received an intravenous infusion of ^{67}Cu -Sar-bisPSMA over about 30 minutes, where the amount (i.e. volume) of the infusion is determined by the dose to be administered to the patient. A second administration of the same dose was provided after at least four weeks to the same patient.

The claims defining the invention are as follows:

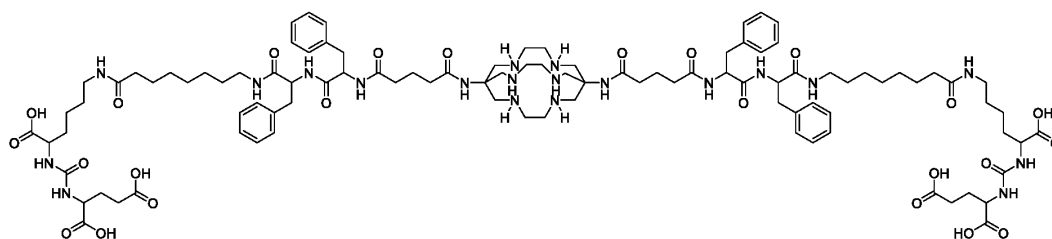
1. A method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:



Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer.

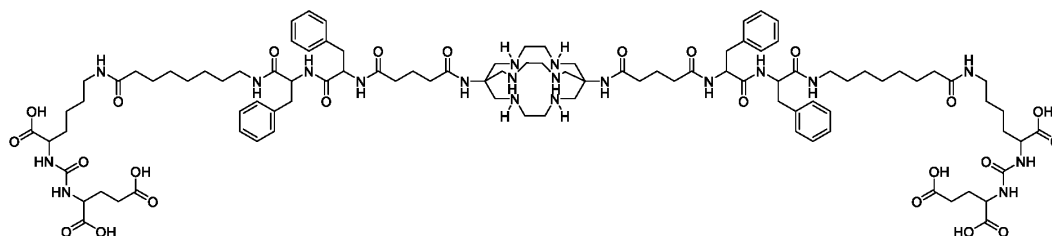
2. A method for the treatment of a cancer, the method comprising administering to a subject in need thereof an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in a therapeutically effective amount:



Formula (I)

wherein the cancer is associated with expression or overexpression of the PSMA membrane protein, and wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer and/or mediate a decrease in PSA levels of the subject.

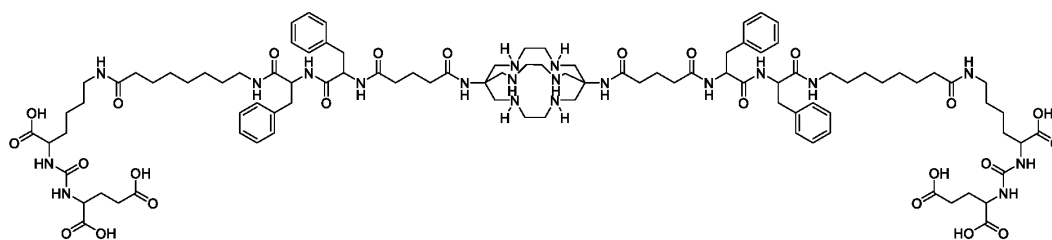
3. A method according to claim 1 or 2, wherein the cancer is a prostate cancer.
4. A method according to claim 3, wherein the cancer is a metastatic prostate cancer.
5. A method according to claim 3 or 4, wherein the prostate cancer is PSMA-expressing metastatic castration resistant prostate cancer (mCRPC).
6. A method for reducing the prostate specific antigen (PSA) level in a subject, the method comprising administering to the subject in need thereof a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount:



Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present.

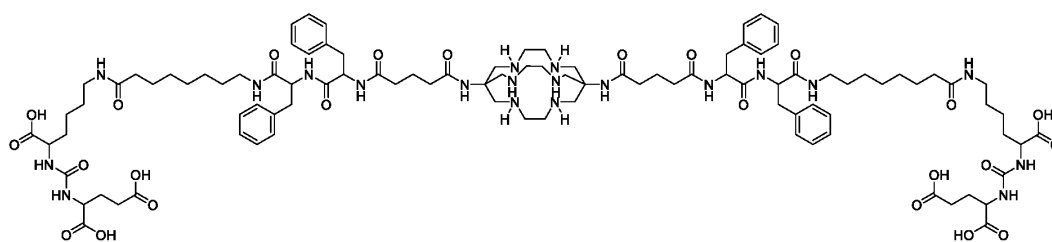
7. A method for reducing the prostate specific antigen (PSA) level in a subject, the method comprising administering to the subject in need thereof a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount:



Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0 ng/ml, wherein the prostate gland of the subject is absent.

8. A method according to any one of claims 1 to 7, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is between about 50 MBq/kg and about 400 MBq/kg.
9. A method according to any one of claims 1 to 8, wherein the method further comprises radioimaging of the subject by PET and/or CT.
10. A method according to any one of claims 1 to 9, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is about 4 GBq, about 8 GBq, about 12 GBq, about 16 GBq, about 20 GBq or about 24 GBq.
11. A method according to any one of claims 1 to 10, wherein the dose is delivered multiple times and that the dose amount may be the same or different.
12. A method according to claim 11, wherein multiple doses are administered between 6 to 16 weeks apart.
13. A method according to any one of claims 1 to 12, wherein the total dose of radiation delivered to the bone marrow of the subject is less than about 2 Gy, the total dose of radiation delivered to the kidneys of the subject is less than about 23 Gy and/or the total dose of radiation delivered to the submandibular gland of the subject is less than about 24 Gy.
14. A method for determining the dose of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope for use in the treatment of a cancer associated with expression or overexpression of the PSMA membrane protein in a subject in need thereof, the method comprising administering to the subject an aqueous formulation of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{64}Cu radioisotope:

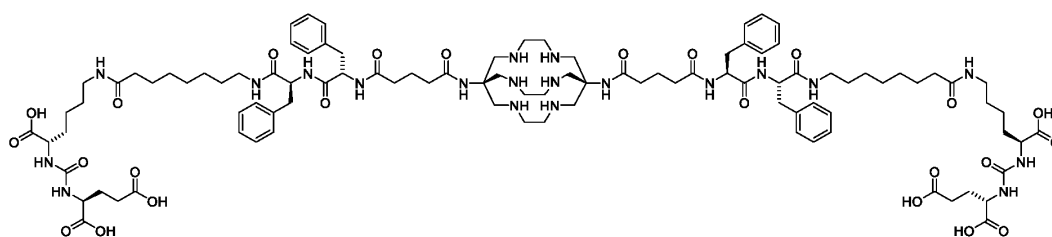


Formula (I)

15. A method according to claim 14, further comprising the step of radioimaging of the subject and determining the uptake of the compound of Formula (I) complexed with a ^{64}Cu radioisotope.

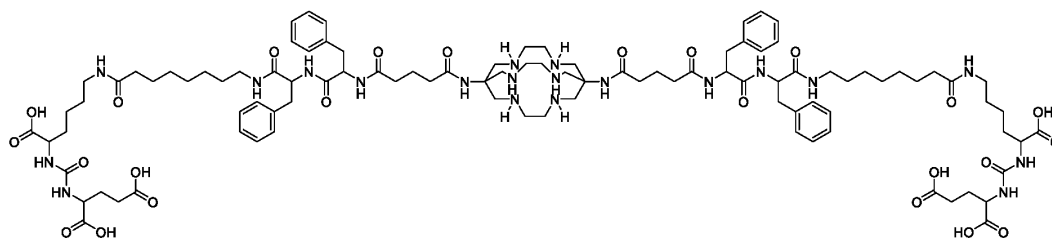
16. A method according to claim 14 or 15, further comprising the step of performing one or more physiological assays of the subject.

17. A method according to any one of claims 1 to 16 wherein the compound of Formula (I) has the structure of Formula (Ia):



Formula (Ia)

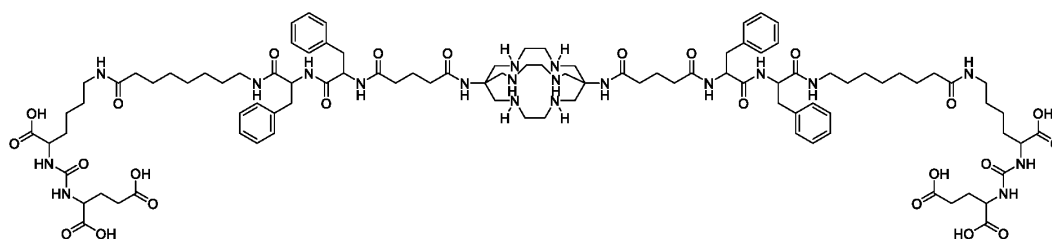
18. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of PSMA membrane antigen, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer.

19. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope:



Formula (I)

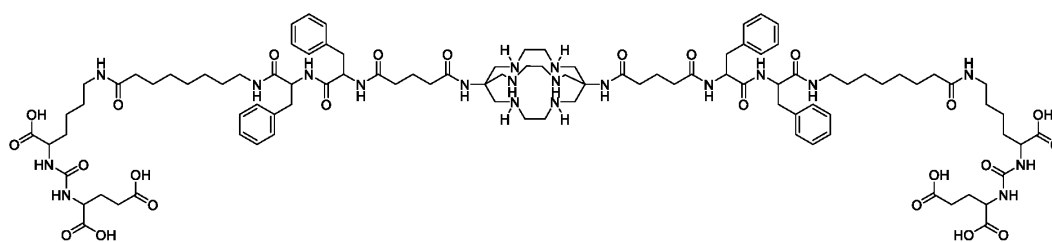
in the manufacture of an aqueous formulation for the treatment of a cancer associated with expression or overexpression of PSMA membrane antigen, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer and/or mediate a decrease in prostate specific antigen (PSA) levels of the subject.

20. Use according to claim 18 or 19, wherein the cancer is a prostate cancer.

21. Use according to claim 20, wherein the cancer is a metastatic prostate cancer.

22. Use according to claim 20 or 21, wherein the prostate cancer is PSMA-expressing metastatic castrate resistant prostate cancer.

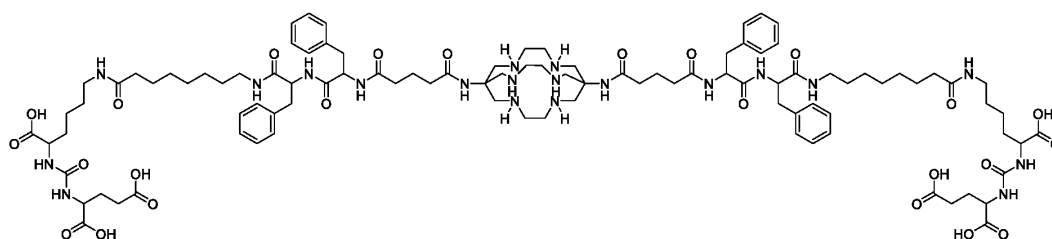
23. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount in the manufacture of an aqueous formulation for reducing prostate specific antigen (PSA) levels in a subject in need thereof:



Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present.

24. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed to a ^{67}Cu radioisotope in an effective amount in the manufacture of an aqueous formulation for reducing prostate specific antigen (PSA) levels in a subject in need thereof,;

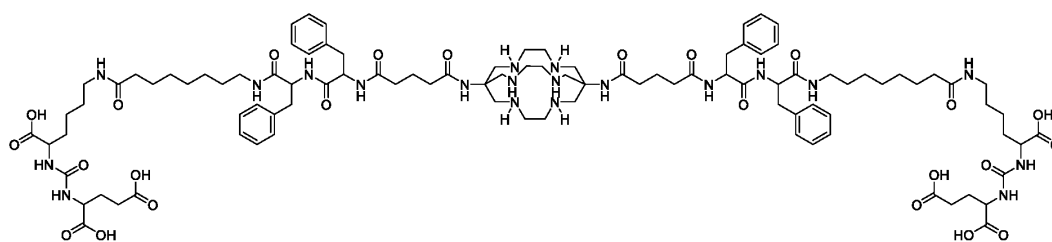


Formula (I)

wherein the PSA levels of the subject prior to administration is greater than about 0 ng/ml, wherein the prostate gland of the subject is absent.

25. Use according to any one of claims 17 to 19, wherein the dose of radiation delivered by the radioisotope is about 4 GBq, about 8 GBq, about 12 GBq, about 16 GBq, about 20 GBq or about 24 GBq.

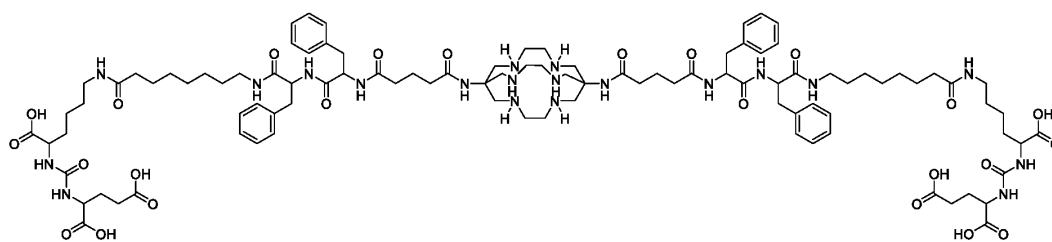
26. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for the treatment of a cancer associated with expression or overexpression of PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer.

27. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

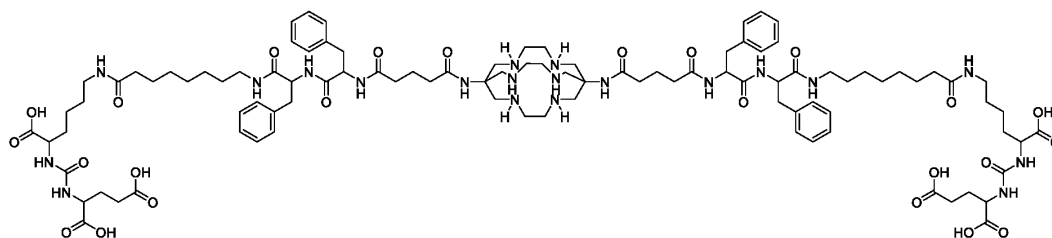
for the treatment of a cancer associated with expression or overexpression of PSMA membrane protein, wherein the dose of radiation delivered by the ^{67}Cu radioisotope is sufficient to reduce the size of one or more lesions associated with the cancer and/or mediate a decrease in prostate specific antigen (PSA) levels.

28. Use according to claim 26 or 27, wherein the cancer is a prostate cancer.

29. Use according to claim 28, wherein the cancer is a metastatic prostate cancer.

30. Use according to claim 28 or 29, wherein the prostate cancer is PSMA-expressing metastatic castrate resistant prostate cancer.

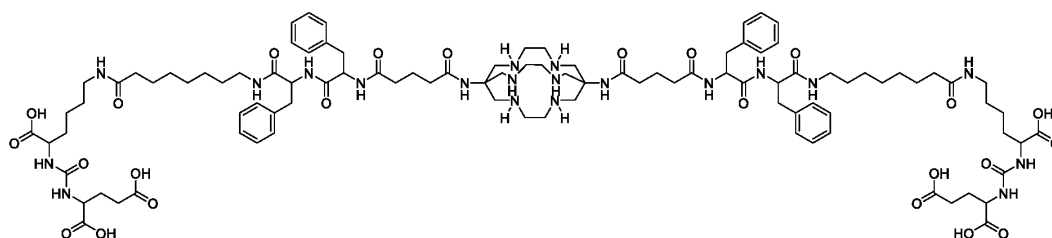
31. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:



Formula (I)

for reducing prostate specific antigen (PSA) levels in a subject in need thereof, wherein the PSA levels of the subject prior to administration is greater than about 0.1 ng/ml, wherein the prostate gland of the subject is present.

32. Use of a compound of Formula (I) or a pharmaceutically acceptable salt thereof complexed with a ^{67}Cu radioisotope:

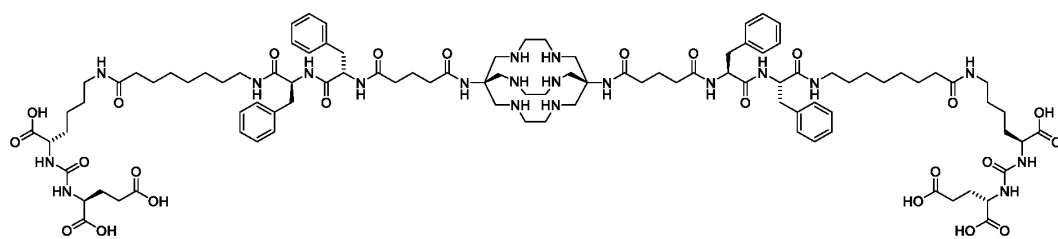


Formula (I)

for reducing prostate specific antigen (PSA) levels in a subject in need thereof, wherein the PSA levels of the subject prior to administration is greater than about 0 ng/ml, wherein the prostate gland of the subject is absent.

33. Use according to any one of claims 26 to 32, wherein the dose of radiation delivered by the radioisotope is about 4 GBq, about 8 GBq, about 12 GBq, about 16GBq, about 20 GBq or about 24 GBq.

34. Use according to any one of claims 26 to 33, wherein the compound of Formula (I) has the structure of Formula (Ia):



Formula (Ia)