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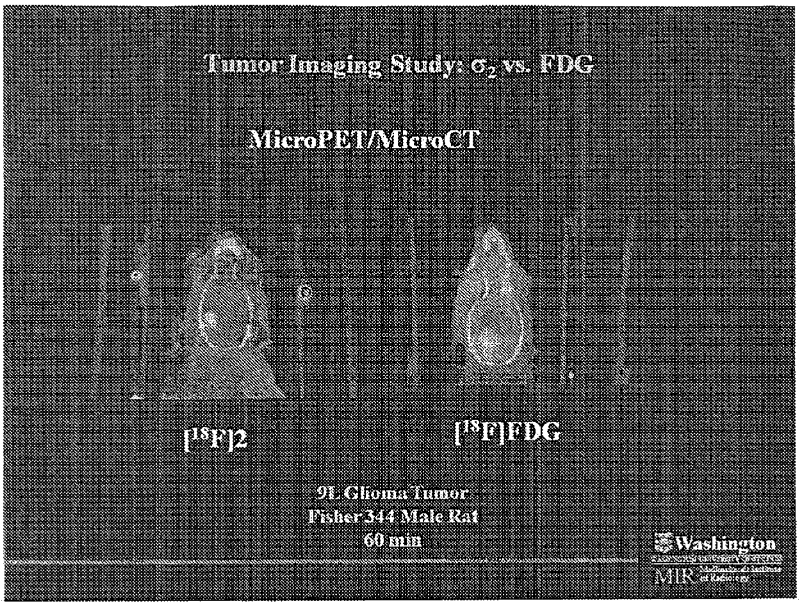

Fig. 9

(57) Abstract: Fluoroalkoxybenzamide compounds which selectively bind Sigma-2 receptors are disclosed. These compounds, when labelled with ^{18}F, can be used as radiotracers for imaging of tumors by positron emission tomography (PET). In addition, these compounds, when labelled with ^{123}I, can be used as radiotracers for imaging of tumors by single photon emission computed tomography (SPECT). Methods for synthesis of these compounds are also disclosed.

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RADIOLABELLED FLUOROBENZAMIDE ANALOGUES, THEIR SYNTHESIS AND USE IN DIAGNOSTIC IMAGING

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority from U.S. Non-Provisional Application Serial No. 11/757,246, filed June 1, 2007, which is a Continuation-in-Part of U.S. Application Serial No. 10/903,771 filed July 30, 2004, which claims priority to U.S. Provisional Application Serial No. 60/491,582 filed July 31, 2003. These applications are hereby incorporated by reference in their entirety.

INTRODUCTION

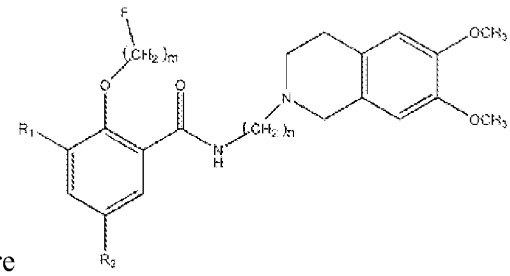
[0002] Sigma receptors are a class of receptors that are expressed in many normal tissues, including liver, kidneys, endocrine glands, and the central nervous system (CNS) (Walker, J.M., et al. Pharmacol Rev 42: 355– 402 1990). It has been well established that there are at least two types of sigma receptors, sigma-1 (σ_1) and sigma-2 (σ_2) (Walker, J.M., et al. Pharmacol Rev 42, 355– 402, 1990). Overexpression of σ_2 receptors has been reported in a variety of human and murine tumors (Bem, W.T., et al., Cancer Res 51: 6558–6562, 1991; Vilner, B.J., et al., In: Multiple sigma and PCP receptor ligands: mechanisms for neuromodulation and neuroprotection?, Kamenka, J.M., and Domino, E.F., ed, Ann Arbor (Mich), 7 NPP Books, p. 341–353, 1992; Mach, R.H., et al., Cancer Res. 57: 156–161, 1997).

[0003] Searches for σ_2 selective ligands has led to the identification of a number compounds having modest to high selectivity for σ_2 versus σ_1 receptors (Fig. 5). These include CB-184 (**10**), CB-64D (**11**), BIMU-1 (**12**) (Bowen, W.D., et al., Eur. J. Pharmacol. 278: 257–260, 1995; Bonhaus, D.W., et al., J. Pharmacol. Exp. Ther. 267: 96, 1993). and PB-167 (**13**) (Colabufo, N. A., et al., J. Pharmacy and Pharmacology 57: 1453-1459, 2005; Kassiou, M., et al., Bioorganic and Medicinal Chemistry, 13: 3623-3626, 2005; Berardi, F., et al., J. Med. Chem. 2004, 47: 2308 – 2317) as well as certain benzamide analogs (**14-16**) (Mach, R.H., et al., Bioorg. Med. Chem. 11: 225, 2003; Huang, Y., et al., J. Med. Chem. 44: 1815, 2001; US Patent application 10/903,771 to Mach et al). We previously reported the evaluation of several ^{11}C, ^{76}Br and $^{125/123}I$ radiolabelled conformationally-flexible benzamide analogs using EMT-6 tumor-bearing female Balb/c mice (Tu, Z., et al., Nucl. Med. Biol. 32: 423-430, 2005; Xu, J., et al., Eur. J. Pharmacol. 21: 525 (1-3): 8-17, 2005; Hou, C., et al., Nucl. Med. Biol. Feb, 33: 203-9, 2006). Initial in vivo studies of 5-methyl-2-[^{11}C]-methoxy-N-[2-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-benzamide and 5-[^{76}Br]-bromo-2,3-dimethoxy-N-[2-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-benzamide indicated that these compounds were potential radiopharmaceuticals for imaging solid tumors and their proliferative status with positron emission tomography (PET). However, the radionuclide properties of ^{76}Br and ^{11}C make these isotopes less than ideal for PET imaging. For example, images produced by PET using ^{76}Br as a radiotracer are often blurry, (Laforest. R., et al., IEEE Transactions on Nuclear Science, 49: 2119-2126, 2002), and the short half-life of ^{11}C ($t_{1/2}$ = 20.4 min) places time constraints on tracer synthesis and duration of scan sessions. Contrast between tumor and normal tissues can be less than satisfactory when a σ_2-selective radiotracer tagged with ^{11}C is used in PET imaging. Accordingly, alternative σ_2-selective ligands for use as radiotracers in PET imaging are needed.

SUMMARY

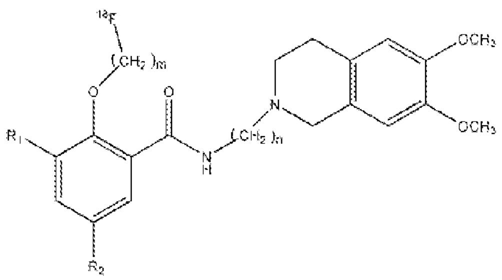
[0004] The present inventors have developed a series of compounds which can be used as radiolabels for diagnostic imaging, in particular positron emission tomography (PET) imaging of tumors. The compounds selectively bind Sigma receptors, and in particular bind Sigma-2 receptors in preference to Sigma-1 receptors. The compounds also selectively bind to tumor cells, and thus can also be used as tracers for detecting tumor cells. In addition, because in some embodiments, the compounds comprise the radioisotope ^{18}F, they can be used as radiotracers for imaging tumors using PET.

[0005] In some embodiments, a tracer the present teachings is a fluoroalkoxybenzamide compound having a

structure 

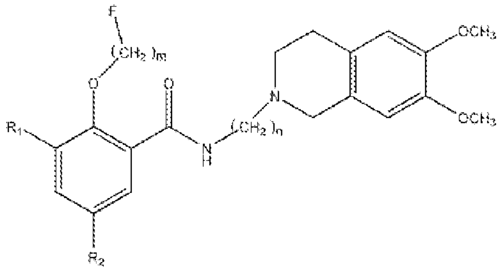
wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof.

[0006] In some embodiments, a compound of the present teachings can include at least one ^{18}F isotope. A compound of these embodiments can be a radiolabelled fluoroalkoxybenzamide compound having a structure

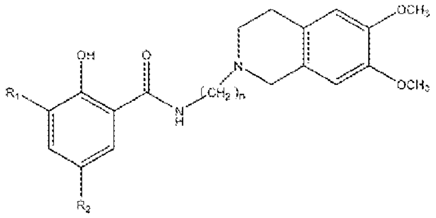


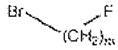
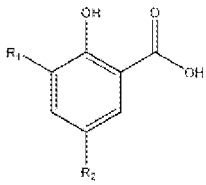
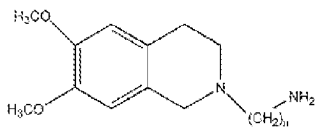
wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof.

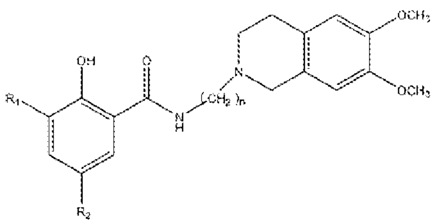
[0007] Other embodiments of the present teachings include methods of synthesizing a fluoroalkoxybenzamide compound of structure



These methods comprise reacting a compound of structure

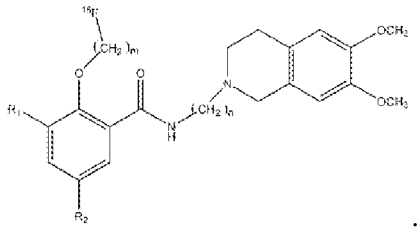


with an fluorinated compound such as , wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2. In some aspects of these embodiments, m can be 2, n can be 2, R_1 can be H, and R_2 can be CH_3. In some aspects, these methods can further comprise reacting  with , thereby forming

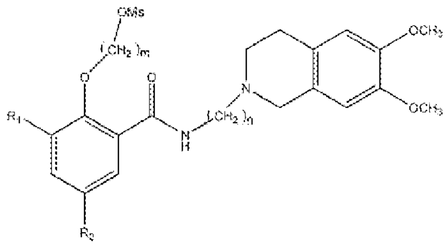


. In other aspects, m can be 2, n can be 4, R_1 can be selected from the group consisting of OCH_3 and H, and R_2 can be selected from the group consisting of Br, CH_3, and I. In yet other aspects, m=2, n=4, R_1 is H, and R_2 is I.

[0008] In yet other embodiments of the present teachings, the inventors disclose methods for synthesizing radiolabelled fluoroalkoxybenzamide compounds of structure

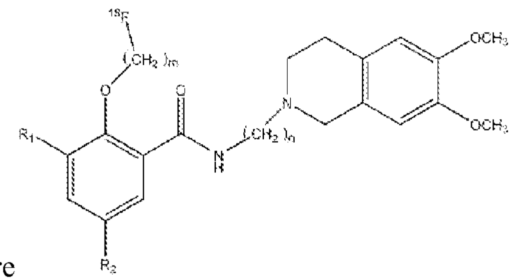


. These methods comprise forming a mixture comprising i) an organic solvent, ii) a compound of structure



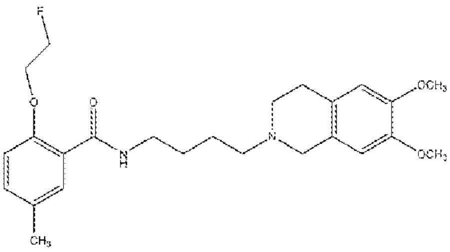
, iii) $^{18}F^-$, iv) 4,7,13,16,21,24-Hexaoxa-1,10-diazabicyclo[8.8.8]-hexacosane and v) a potassium salt, wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2. In some aspects of these methods, m can be 2, n can be 4, R_1 can be selected from the group consisting of H and OCH_3, and R_2 can be selected from the group consisting of CH_3, Br and I. In yet other aspects of these methods, m=2, n=4, R_1 is OCH_3, and R_2 is I. In addition, in various aspects, the potassium salt can be K_2CO_3, and the organic solvent can be dimethyl sulfoxide, acetonitrile or a combination thereof. Furthermore, in various aspects, the methods can include heating a mixture.

[0009] In additional embodiments of the present teachings, the inventors disclose methods of imaging a tumor in a mammal such as a human. These methods comprise administering to the mammal a radiolabelled fluoroalkoxybenzamide compound of structure



wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof; and subjecting the mammal to positron emission tomography (PET) scanning. In some aspects, m can be 2, n can be 4, R_1 can be selected from the group consisting of H and OCH_3, and R_2 can be selected from the group consisting of CH_3, Br and I.

[0010] In various aspects of the above embodiments, a fluoroalkoxybenzamide compound or a salt thereof can include particular molecular species, such as,



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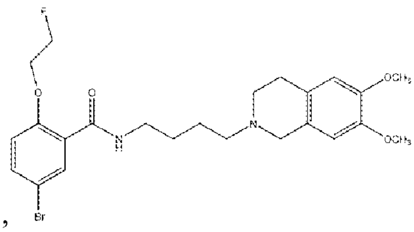

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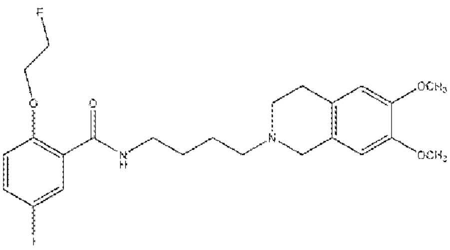

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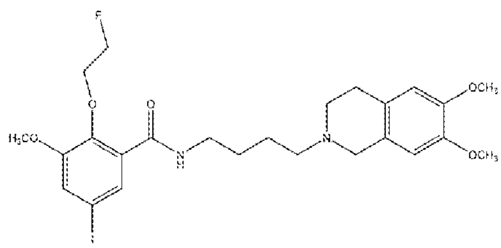
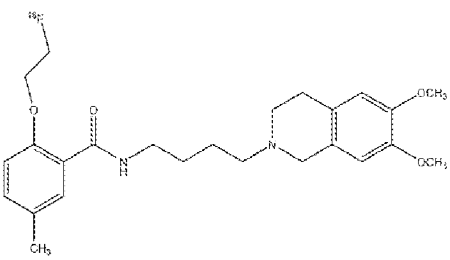


or salts thereof.

[0011] In various aspects of these embodiments, a radiolabelled fluoroalkoxybenzamide compound or a salt thereof can include particular molecular species, such as,



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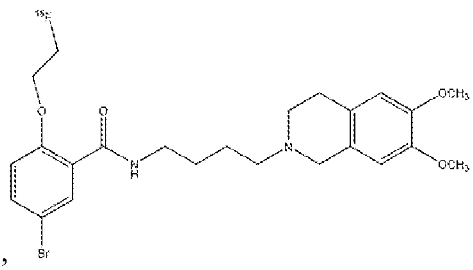

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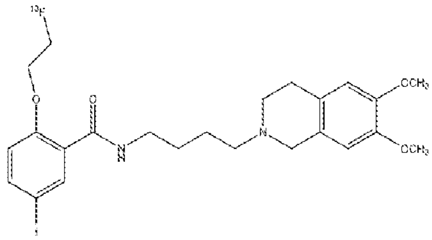
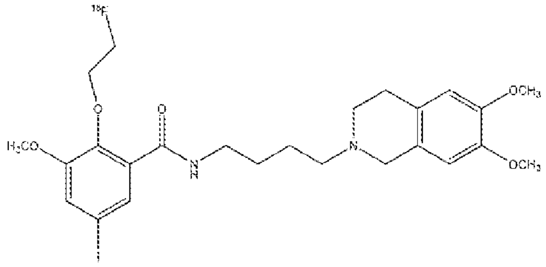
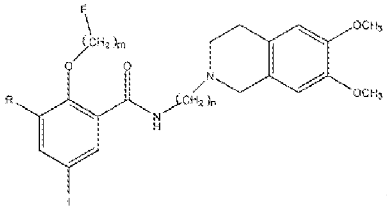
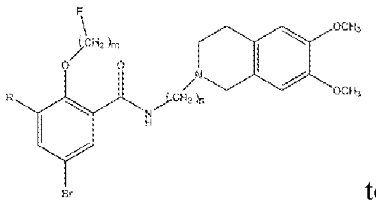
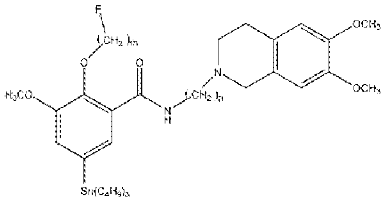
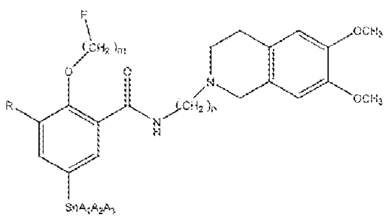
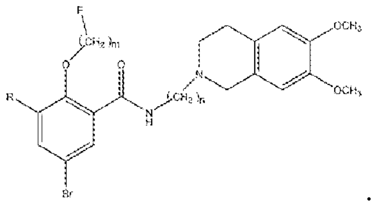
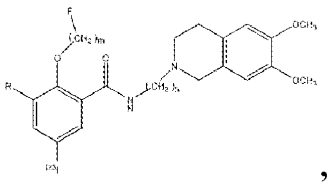
 ,  or salts thereof.

[0012] In additional embodiments of the present teachings, the inventors disclose methods for synthesizing compounds of structure  wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R is H or a C_{1-4} alkoxy such as a methoxy. These methods comprise:

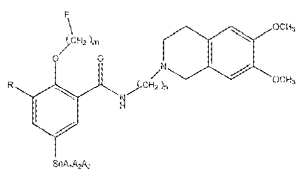
stannylating a compound of structure  to form a compound of structure  and

iodinating a compound of structure  . In some configurations, the stannylated compound can be formed by stannylating a compound of structure  . In some aspects of these methods, m can be 2 and n can be 4.

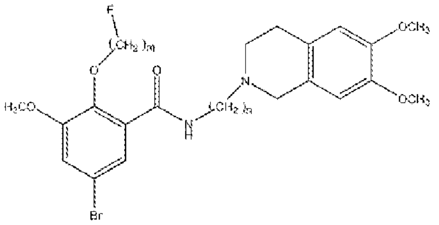
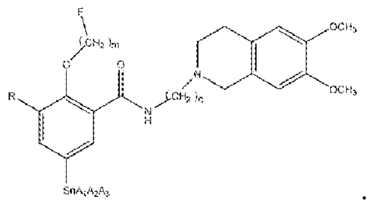
[0013] In yet other embodiments of the present teachings, the inventors disclose iodine-123 radiolabelled fluoroalkoxybenzamide compounds of structure



, wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R can be H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, and salts thereof. In various configurations, m can be 2, n can be 4, R can be H or a C_{1-4} alkoxy such as a methoxy.

[0014] In related embodiments, the inventors disclose methods for synthesizing these radioiodinated compounds. In various configurations, these methods include reacting a compound of structure 
with [^{123}I]NaI and an oxidant, wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, A_1, A_2 and A_3 are each independently a C_{1-4} alkyl, and R is selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2. In various aspects, R can be H or a C_{1-4} alkoxy such as a methoxy; A_1, A_2 and A_3 can each be independently a butyl moiety selected from an n-butyl moiety, an iso-butyl moiety, a

sec-butyl moiety, and a tert-butyl moiety. In some configurations, A_1, A_2 and A_3 can each be an n-butyl moiety, m can be 2 and n can be 4. In addition, in various aspects, the oxidant can be peracetic acid, hydrogen peroxide, chloramine T (*N*-chloro-*p*-toluenesulfonamide sodium salt) or a combination thereof.

[0015] In some aspects of these embodiments, the methods can further include stannylating a compound of structure  to form a compound of structure .

[0016] In yet other embodiments, the present teachings include methods of imaging a solid tumor in a mammal such as a human. In various configurations, these methods include administering to the mammal a radioiodinated compound described above, and subjecting the mammal to single photon emission computed tomography (SPECT) imaging.

BRIEF DESCRIPTION OF THE DRAWINGS

Fig. 1 illustrates scheme I for synthesis of some compounds of the present teachings.

Fig. 2 illustrates scheme II for synthesis of some compounds of the present teachings.

Fig. 3 illustrates scheme III for synthesis of some compounds of the present teachings.

Fig. 4 illustrates scheme IV for synthesis of some compounds of the present teachings.

Fig. 5 illustrates structure and properties of several σ_2 selective ligands.

Fig. 6 illustrates tumor : organ ratios for the ^{18}F-labeled σ_2 selective ligands, **3c-f**, at 1 h (top) and 2 h (bottom) after i.v. injection into female Balb/c mice bearing EMT-6 tumors.

Figure 7 presents a comparison of the tumor : fat and tumor : muscle ratios for **[^{18}F]3c** or **[^{18}F]3f** when there is no-carrier-added and when the σ_1 and σ_2 receptors are blocked with 1 mg/kg of YUN-143. All values were obtained 1 h after injection of the radiotracer.

Figure 8 illustrates microPET and microCT images of EMT-6 tumors in female Balb/c mice. All MicroPET images were acquired 1 h after i.v. injection of either **[^{18}F]3c** or **[^{18}F]3f**.

Fig. 9 illustrates an image of a glioma using [^{18}F]3f of the present teachings compared to [^{18}F]FDG.

DETAILED DESCRIPTION

[0017] The present inventors have developed a series of compounds which can be used as radiolabels for diagnostic imaging, in particular positron emission tomography (PET) imaging of tumors. The compounds selectively bind Sigma receptors, and in particular bind Sigma-2 receptors in preference to Sigma-1 receptors. The compounds also selectively bind to tumor cells, and thus can be used as tracers for detecting tumor cells. Without being limited by theory, it is generally believed that many types of tumor cells have a high density of sigma-2 receptors, and therefore compounds of the present teachings are effective tracers for detecting tumors by virtue of the compounds' affinity for the sigma-2 receptors. In addition, because in some embodiments, the compounds comprise the radioisotope ^{18}F, a preferred isotope for imaging by positron emission tomography (PET) they are effective as radiotracers for PET imaging of tumors in humans or other mammals. Furthermore, in some embodiments, the compounds comprise the radioisotope ^{123}I, a preferred isotope for imaging by single photon emission computed tomography (SPECT). These compounds are effective as radiotracers for SPECT imaging of tumors in humans or other mammals.

[0018] The present inventors have synthesized several novel conformationally flexible benzamide analogues having a moderate to high binding affinity and selectivity for σ_2 receptors (Table I). Four of these compounds were selected as candidates for developing ^{18}F-labeled PET probes to image the σ_2 receptor status of solid tumors. **[^{18}F]3c, [^{18}F]3d, [^{18}F]3e,** and **[^{18}F]3f,** were successfully synthesized and evaluated as potential radiotracers for imaging EMT-6 tumors in female Balb/c mice. Of the four ^{18}F-labeled analogues, **[^{18}F]3c** and **[^{18}F]3f** had the best biodistribution kinetics and tumor : normal tissue ratios. Blocking studies confirmed that the uptake of **[^{18}F]3c** and **[^{18}F]3f** was σ_2-receptor mediated. Our studies indicate that various compounds of the present teachings, including **[^{18}F]3c** and **[^{18}F]3f,** are acceptable agents for detecting and imaging solid tumors and their σ_2 receptor status with PET.

[0019] The present inventors devised a design strategy for generating σ_2-selective ligands of the present teachings. This strategy involved replacing the ortho methoxy group of ^{11}C-labelled benzamide analogs (Xu, J., et al., Eur. J. Pharmacol. 21;525 (1-3): 8-17, 2005) with a 2-fluoroethyl group as shown in Scheme I (Fig. 1) and in Examples below.

[0020] The following examples are illustrative of the various embodiments of the present teachings. The examples are not intended to limit the scope of the claims. The methods described herein utilize laboratory techniques well known to skilled artisans, and guidance can be found in laboratory manuals and textbooks such as Sambrook, J., et al., Molecular Cloning: A Laboratory Manual, 3rd ed. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 2001; Spector, D. L. et al., Cells: A Laboratory Manual, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1998; and Harlow, E., Using Antibodies: A Laboratory Manual, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1999; Hedrickson et al., Organic Chemistry 3rd edition, McGraw Hill, New York, 1970; Carruthers, W., and Coldham, I., Modern Methods of Organic Synthesis (4th Edition), Cambridge University Press, Cambridge, U.K., 2004; Curati, W.L., Imaging in

Oncology, Cambridge University Press, Cambridge, U.K., 1998; Welch, M.J., and Redvanly, C.S., eds. Handbook of Radiopharmaceuticals: Radiochemistry and Applications, J. Wiley, New York, 2003.

[0021] In the experiments described in herein, all reagents were purchased from commercial suppliers and used without further purification unless otherwise stated. Tetrahydrofuran (THF) was distilled from sodium hydride immediately prior to use. Anhydrous toluene was distilled from sodium/toluene shortly before use. All anhydrous reactions were carried out in oven-dried glassware under an inert nitrogen atmosphere unless otherwise stated. When the reactions involved extraction with dichloromethane (CH_2Cl_2), chloroform ($CHCl_3$), ethyl acetate (EtOAc), or ethyl ether (Et_2O), the organic solutions were dried with anhydrous Na_2SO_4 and concentrated with a rotary evaporator under reduced pressure. Flash column chromatography was conducted using silica gel 60a, "40 Micron Flash" [32-63μm] (Scientific Adsorbents, Inc.). Melting points were determined using the MEL-TEMP 3.0 apparatus and left uncorrected. ^1H NMR spectra were recorded at 300 MHz on a Varian Mercury-VX spectrometer with $CDCl_3$ as solvent and tetramethylsilane (TMS) as the internal standard. All chemical shift values are reported in ppm (□). Elemental analyses (C, H, N) were determined by Atlantic Microlab, Inc.

EXAMPLE 1

[0022] This example demonstrates reactions yielding fluoroalkoxy 2-hydroxybenzamide analogs. As illustrated in Fig. 1, Scheme I involves condensation of compounds **1a** and **1b** with a substituted salicylic acid to give the corresponding substituted 2-hydroxybenzamide analogs, **2a-e.** Alkylation of the ortho hydroxyl group with 2-bromo-1-fluoroethane using potassium carbonate as a base produced **3a-e** in moderate to high yield. Compound **3f** was prepared by iodination of the corresponding tin precursor, **3g,** which was prepared from **3b** using standard stannylation reaction conditions. Compounds, **3a-f,** were then converted into either the hydrochloride or oxalic acid salts for the in vitro σ_1 and σ_2 receptor binding assays.

EXAMPLE 2

[0023] This example illustrates synthetic steps for generating ^{18}F-tagged compounds of the present teachings. In this example, compounds **3c-f** were radiolabeled with ^{18}F as shown in Schemes II-IV (Figs. 2, 3 and 4, respectively). Scheme II (Fig. 2) outlines the synthesis of the mesylate precursors required for the radiolabeling procedure. Alkylation of the ortho hydroxyl group of compounds, **2c-e,** with 1-bromoethyl acetate followed by hydrolysis of the acetate group produced the corresponding 2-hydroxyethyl analogs, **4c-e,** in good yield. Compounds, **4c-e,** were then converted to the corresponding mesylates, **5c-e,** by treatment with methanesulfonyl chloride in dichloromethane using triethylamine as an acid scavenger.

EXAMPLE 3

[0024] This example illustrates synthesis of the precursor for the corresponding 5-iodo analog, **5f,** as shown in Scheme III (Fig. 3). Esterification of 5-bromo-2-methoxy salicylic acid followed by alkylation of the ortho hydroxyl group with 1-bromoethyl acetate, then hydrolysis of the acetate and benzoate esters produced the corresponding 2-hydroxyethyl analog, **9**. Condensation of **9** with the amine, **1b**, gave the amide, **4f**, which was converted to the corresponding mesylate, **5f**, using the conditions described above for the analogs, **5c-e**.

EXAMPLE 4

[0025] This example illustrates synthesis of **[^{18}F]3c, [^{18}F]3d, [^{18}F]3e,** and **[^{18}F]3f** from mesylate precursors. As shown in Scheme IV (Fig. 4), mesylate precursors, compounds **5c-f, were treated** with [^{18}F]fluoride/potassium carbonate and 4,7,13,16,21,24-Hexaoxa-

1,10-diazabicyclo[8.8.8]-hexacosane (Kryptofix 222®, Acros Organics N.V., Fairlawn, NJ) using dimethyl sulfoxide (DMSO) as the solvent. The reaction mixture was irradiated for 30-40 seconds in a microwave oven, and the crude product separated from the unreacted [^{18}F]fluoride using a C-18 reverse phase Sep-Pak® cartridge (Waters Corp., Milford, MA) and methanol as the eluant. The crude product was then purified by high-performance liquid chromatography (HPLC) using a C-18 reverse phase column. The entire procedure required ~2 h, and the radiochemical yield, corrected for decay to the start of synthesis, was 20~30%. The specific activities ranged from 1500 - 2500 Ci/mmol.

EXAMPLE 5

[0026] This example illustrates in vitro binding studies with the compounds of the present teachings. In this example, in vitro binding studies were conducted in order to measure the affinity of the target compounds for σ_1 and σ_2 receptors.

[0027] In these assays, the novel sigma ligands were dissolved in *N,N*-dimethylformamide (DMF), DMSO or ethanol, and then diluted in 50 mM Tris-HCl buffer containing 150 mM NaCl and 100 mM EDTA at pH 7.4 prior to performing the σ_1 and σ_2 receptor binding assays The procedures for isolating the membrane homogenates and performing the σ_1 and σ_2 receptor binding assays have been described in detail previously (Xu, J., et al., Eur. J. Pharmacol. 21: 525 (1-3): 8-17, 2005. Briefly, the σ_1 receptor binding assays were conducted in 96-well plates using guinea pig brain membrane homogenates (~300 μg protein) and ~5 nM [^{3}H](+)-pentazocine (34.9 Ci/mmol, Perkin Elmer, Boston, MA). The total incubation time was 90 min at room temperature. Nonspecific binding was determined from samples that contained 10 μM of cold haloperidol. After 90 min, the reaction was terminated by the addition of 150 μL of ice-cold wash buffer (10 mM Tris-HCl, 150 mM NaCl, pH 7.4) using a 96 channel transfer pipette (Fisher Scientific, Pittsburgh, PA). The samples were harvested and filtered rapidly through a 96-well fiber glass filter plate

(Millipore, Billerica, MA) that had been presoaked with 100 μL of 50 mM Tris-HCl buffer at pH 8.0 for 1 h. Each filter was washed 3 times with 200 μL of ice-cold wash buffer, and the filter counted in a Wallac 1450 MicroBeta liquid scintillation counter (Perkin Elmer, Boston, MA).

[0028] The σ_2 receptor binding assays were conducted using rat liver membrane homogenates (~300 μg protein) and ~5 nM [^{3}H]DTG (58.1 Ci/mmol, Perkin Elmer, Boston, MA) in the presence of 1 μM (+)-pentazocine to block σ_1 sites. The incubation time was 120 min at room temperature. Nonspecific binding was determined from samples that contained 10 μM of cold haloperidol. All other procedures were identical to those described for the σ_1 receptor binding assay above.

[0029] Data from the competitive inhibition experiments were modeled using nonlinear regression analysis to determine the concentration that inhibits 50% of the specific binding of the radioligand (IC_{50} value). Competitive curves were best fit to a one-site fit and gave pseudo-Hill coefficients of 0.6 – 1.0. K_i values were calculated using the method of Cheng and Prusoff (Biochem. Pharmacol. 22: 3099-3108, 1973) and are presented as the mean ± 1 SEM. For these calculations, we used a K_d value of 7.89 nM for [^{3}H](+)-pentazocine and guinea pig brain; for [^{3}H]DTG and rat liver, we used 30.73 nM [20]

[0030] The binding assays used [^{3}H](+)-pentazocine for the σ_1 receptors and [^{3}H]1 ,3-Di(2-tolyl)guanidine ([^{3}H]DTG) in the presence of 100 nM (+)-pentazocine for the σ_2 receptors. The K_i values were determine from Scatchard plots. The results of the binding assays for compounds, **3a-f,** are shown in Table I. Increasing the length of the spacer group from two carbons (**3a**) to 4 carbons (**3c**) results in a 69-fold increase in the affinity for σ_1 receptors, a 15-fold increase in the affinity for σ_2 receptors, and a 0.5 unit increase in the log D value; a measure of the lipophilicity of the compounds (Table I). Although four of the five compounds (**3c-f)** with a four-carbon spacer had higher affinities for σ_1 receptors (their K_i values ranged from 330 to 2,150 nM) than the compound (**3a**) with a two-carbon spacer (K_i =

22,750 nM), the affinities of **3c-f** for σ_2 receptors increased proportionately more (their K_i values ranged from 0.26 to 6.95 nM), leading to substantial increases in their $\sigma_2 : \sigma_1$ ratios (Table I).

[0031] The σ_2:σ_1 ratios for compounds, **3c-f,** varied from 48 to 8,190. The excellent σ_2 receptor affinities and moderate to high σ_2:σ_1 ratios for the compounds, **3c-f,** indicated that their corresponding ^{18}F-labeled analogs would be useful radiotracers for imaging the σ_2 receptor status of solid tumors with PET. Also, the log D values for these compounds, a measure of their lipophilicity, are within the range that should lead to a high uptake in solid tumors (Xu, J., et al., Eur. J. Pharmacol. 21: 525 (1-3): 8-17, 2005).

Table I. Affinity (K_i) of the benzamide analogs**,** 6a-e, for the σ_1 and σ_2 receptors assayed in vitro

	K_i value (nM)			
	σ_1	σ_2	σ_1:σ_2 Ratio	Log D[a] (pH=7.4)
3a	22,750 ± 3,410	102 ± 4	222	2.54
3b	15,300 ± 2,305	386 ± 93	40	3.56
3c	330 ± 25	6.95 ± 1.63	48	3.06
3d	1,076 ± 88	0.65 ± 0.22	1,656	3.89
3e	1,300 ± 225	1.06 ± 0.30	1,230	4.13
3f	2,150 ± 410	0.26 ± 0.07	8,190	3.46

[a]calculated using the program ACD/log D

EXAMPLE 6

[0032] This example illustrates in vivo evaluation of compounds of the present teachings. All animal experiments were conducted in compliance with the Guidelines for the Care and Use of Research Animals established by Washington University's Animal Studies Committee. EMT-6 mouse mammary adenocarcinoma cells (5 x10^5 cells in 100 uL of phosphate-buffered saline) were implanted subcutaneously in the scapular region of female

Balb/c mice (~2-month old and 17–22 g; Charles River Laboratories). The biodistribution studies were initiated 7-10 days after implantation when the tumor size was ~0.2 cm^3 (~200 mg).

[0033] For the biodistribution studies, 10– 120 µCi of **[^{18}F]3c, [^{18}F]3d, [^{18}F]3e** or **[^{18}F]3f** in 100–150 uL of saline was injected via the tail vein into EMT-6 tumor-bearing female Balb/c mice. Groups of at least 4 mice were used for each time point. At 5, 30, 60, and 120 min after injection, the mice were euthanized, and samples of blood, lung, liver, kidney, muscle, fat, heart, brain, bone and tumor were removed, weighed and counted in a Beckman Gamma 8000 well counter. After counting, the percentage of the injected dose per gram of tissue (%ID/g) was calculated. The tumor/organ ratios were calculated by dividing the %ID/g of the tumor by the %ID/g of each organ.

[0034] The results of the biodistribution studies in female Balb/c mice bearing EMT-6 tumors are shown in Table II. All four labeled compounds displayed excellent tumor uptake at 5 min post-injection, with values ranging from 2.5-3.7 percent of the injected dose per gram (%ID/g). Tumor uptake at 1 h post-injection remained high for each of the ligands, **[^{18}F]3c**, **[^{18}F]3d**, **[^{18}F]3e** and **[^{18}F]3f,** (1.14, 2.09, 2.72, and 2.15 %ID/g, respectively)**,** and continued to remain relatively high at 2 h post-injection (0.64, 0.96, 1.92 and 1.15 %ID/g, respectively) compared to that of the normal tissues, fat and muscle. This resulted in acceptable tumor : normal tissue ratios for the PET imaging studies. For example, the tumor : muscle ratios ranged from 3 – 4 and the tumor : fat ratios ranged from 4.5 – 8 at 2 hrs post-injection, respectively. Also, the low bone uptake of all four labeled compounds, which continued to decrease between the 30 min and 1 h time points, suggests that these compounds do not undergo a significant defluorination in vivo.

[0035] Compound **[^{18}F]3f** had the highest tumor : muscle ratio (~8) and a tumor : fat ratio of ~7 at 2 h after i.v. injection (**Figure 6**). The tumor : fat ratios for **[^{18}F]3c** and **[^{18}F]3d** were also high, reaching ~ 8 and ~6, respectively, at 2 h after i.v. injection However, the tumor : muscle ratios for **[^{18}F]3c** and **[^{18}F]3d** were much lower than that for **[^{18}F]3f**. Although the tumor uptake of **[^{18}F]3d** and **[^{18}F]3e** is higher than that of **[^{18}F]3c** at both 1 h and 2 h post-injection, these radiotracers cleared much more slowly from the blood than **[^{18}F]3c** (**Table II),** making them less desirable than **[^{18}F]3c** as PET imaging agents . The moderate to high tumor : normal tissue ratios and the rapid clearance from the blood for **[^{18}F]3c** and **[^{18}F]3f** suggests that these radiotracers are likely the best candidates for imaging of solid tumors with PET. Consequently, these two radiotracers were selected for further studies to evaluate the suitability for detecting solid tumors and imaging their σ_2 receptor status with PET.

Table II. [18F]3c-f Biodistribution in female Balb/c mice bearing EMT-6 tumors

	5 min.	30 min.	60 min.	120 min.	5 min.	30 min.	60 min.	120 min.
		[18F]3c				**[18F]3d**		
blood	2.49 ± 0.49	1.16 ± 0.10	0.56 ± 0.08	0.35 ± 0.05	3.57 ± 0.43	2.81 ± 0.32	1.69 ± 0.63	0.52 ± 0.10
lung	10.26 ± 0.71	2.36 ± 0.19	0.88 ± 0.12	0.43 ± 0.07	12.08 ± 1.98	3.08 ± 0.23	1.60 ± 0.27	0.51 ± 0.08
liver	23.33 ± 4.22	10.51 ± 0.87	4.14 ± 0.55	2.05 ± 0.43	32.60 ± 3.96	13.12 ± 1.39	5.69 ± 0.54	2.30 ± 0.33
kidney	29.18 ± 1.92	6.86 ± 0.45	2.51 ± 0.51	0.87 ± 0.13	42.94 ± 3.34	17.55 ± 2.75	6.92 ± 1.61	1.12 ± 0.16
muscle	1.86 ± 0.08	0.70 ± 0.19	0.34 ± 0.05	0.24 ± 0.08	1.95 ± 0.19	0.98 ± 0.18	0.58 ± 0.11	0.28 ± 0.10
fat	1.95 ± 0.33	0.59 ± 0.13	0.22 ± 0.04	0.08 ± 0.02	2.85 ± 0.47	0.96 ± 0.13	0.38 ± 0.05	0.15 ± 0.06
heart	3.73 ± 0.15	1.15 ± 0.06	0.55 ± 0.07	0.27 ± 0.04	3.74 ± 0.37	1.55 ± 0.11	1.04 ± 0.21	0.40 ± 0.07
brain	0.76 ± 0.06	0.27 ± 0.05	0.18 ± 0.03	0.12 ± 0.02	1.09 ± 0.11	0.40 ± 0.03	0.32 ± 0.05	0.20 ± 0.03
bone	2.49 ± 0.19	0.96 ± 0.15	0.55 ± 0.07	0.45 ± 0.11	2.90 ± 0.39	1.17 ± 0.06	1.12 ± 0.16	1.28 ± 0.28
tumor	3.67 ± 0.45	2.54 ± 0.27	1.14 ± 0.10	0.64 ± 0.10	3.28 ± 0.41	2.59 ± 0.19	2.09 ± 0.28	0.96 ± 0.24
		[18F]3e				**[18F]3f**		
blood	4.60 ± 0.44	4.30 ± 0.59	3.39 ± 0.29	1.92 ± 0.59	1.82 ± 0.25	1.23 ± 0.28	0.65 ± 0.09	0.28 ± 0.01
lung	9.71 ± 0.83	4.07 ± 0.46	2.34 ± 0.12	1.36 ± 0.24	18.47 ± 3.07	3.75 ± 0.58	1.51 ± 0.13	0.74 ± 0.03
liver	37.26 ± 4.88	17.35 ± 2.72	7.31 ± 0.98	4.25 ± 1.56	15.21 ± 2.21	10.73 ± 2.98	5.57 ± 0.31	2.61 ± 0.69
kidney	36.07 ± 2.28	17.43 ± 1.95	9.36 ± 0.90	3.92 ± 0.98	19.98 ± 1.66	7.73 ± 2.25	3.50 ± 0.80	1.34 ± 0.10
muscle	1.52 ± 0.10	1.12 ± 0.05	0.83 ± 0.04	0.60 ± 0.11	2.50 ± 0.33	0.73 ± 0.14	0.40 ± 0.07	0.15 ± 0.01
fat	2.32 ± 0.46	1.04 ±	0.62 ±	0.44 ±	4.13 ±	1.36 ±	0.47 ±	0.17 ±

		0.06	0.10	0.08	0.86	0.50	0.07	0.05
heart	3.22 ± 0.27	2.37 ± 0.29	1.61 ± 0.11	1.00 ± 0.23	5.60 ± 0.45	1.54 ± 0.31	0.69 ± 0.06	0.39 ± 0.04
brain	0.55 ± 0.04	0.44 ± 0.04	0.36 ± 0.02	0.37 ± 0.06	0.71 ± 0.09	0.27 ± 0.04	0.14 ± 0.02	0.08 ± 0.01
bone	2.59 ± 0.28	1.31 ± 0.14	0.99 ± 0.07	1.67 ± 0.27	2.23 ± 0.56	2.01 ± 0.53	0.93 ± 0.16	0.59 ± 0.09
tumor	2.54 ± 0.62	2.81 ± 0.62	2.72 ± 0.13	1.92 ± 0.10	3.05 ± 0.43	3.11 ± 0.16	2.15 ± 0.25	1.15 ± 0.23

EXAMPLE 7

[0036] This example illustrates specificity of binding in vivo for σ_2 receptors by compounds of the present teachings.

[0037]In order to demonstrate that the in vivo binding of **[^{18}F]3c** and **[^{18}F]3f** is specific for σ_2 receptors, a no-carrier-added dose of these radiotracers was co-injected into EMT-6 tumor-bearing mice with *N*-(4-fluorobenzyl)piperidinyl-4-(3-bromophenyl) acetamide (YUN-143), a sigma ligand displaying a high affinity for both σ_1 and σ_2 receptors. Co-injection of YUN-143 with either **[^{18}F]3c** or **[^{18}F]3f** resulted in a significant decrease (~50%) in the tumor : muscle and tumor : fat ratios at 1 h post-injection (Fig. 7).

[0038] These blocking studies in tumor-bearing mice were conducted by co-injecting 1 mg/kg of cold N-(4-fluorobenzyl) piperidinyl-4-(3-bromophenyl)acetamide (YUN-143) with **[^{18}F]3c** or **[^{18}F]3f.** Yun-143 has a high affinity for both σ_1 and σ_2 receptors and is routinely used for sigma receptor blocking studies (Mach, R.H., et al., Nucl Med Biol. 28: 451–458, 2001; Bowen, W.D. et al., Eur. J. Pharmacol. 278: 257–260, 1995; Bonhaus, D.W. et al., J. Pharmacol. Exp. Ther. 267: 961, 1993. All mice were sacrificed 60 min after injection of the radiotracer, and the tumor : organ ratios were determined as described above. The data presented in Fig. 7 indicate that both **[^{18}F]3c** and **[^{18}F]3f** bind selectively to σ_2 receptors in vivo.

EXAMPLE 8

[0039] This example illustrates use of radioligands of the present teachings as imaging agents.

[0040] To confirm the feasibility of using radioligands of the present teachings as PET imaging agents for determining the σ_2 receptor status of solid tumors, a CT/PET study using either **[^{18}F]3c** or **[^{18}F]3f** in female Balb/c mice bearing EMT-6 tumors was performed on a microPET-F220 (CTI-Concorde Microsystems Inc.) and a MicroCAT-II system (ImTek Inc.). For the microPET studies, each mouse was injected with ~0.25 mCi of either **[^{18}F]3c** or **[^{18}F]3f** via the tail vein and imaged 1 h later. MicroCT images were also obtained and co-registered with the PET images to determine the exact anatomical location of the radiotracers.

[0041] In these studies, the EMT-6 tumors were readily identifiable using either radioligand, indicating that they are both acceptable agents for detecting solid tumors and imaging their σ_2 receptor status with PET (Fig. 8).

EXAMPLE 9

[0042] This example describes a general method for synthesis of the substituted 2-hydroxybenzoic acid amides, compounds 2a-e, in particular compound 2a.

[0043] To synthesize N-[2-(6,7-dimethoxy-3,4-dihydro-1*H*-isoquinolin-2-yl)-ethyl]-2-hydroxy-5-methyl-benzamide (compound 2a), 1,3-dicyclohexycarbodimide (432.6 mg, 2.1 mmol) and 1-hydroxybenzotriazole (283.8 mg, 2.10 mmol) were added to a ice-water bath cooled solution of **1a** (472.0 mg, 2.0 mmol) and 2-hydroxy-5-methyl-benzoic acid (152 mg, 2.0 mmol) in 30 ml dichloromethane. After the reaction mixture was stirred overnight, analysis of the products using thin layer chromatography with 20% methanol and 80% ethyl ether as the mobile phase indicated that the reaction was complete. After completion of the reaction, another 50 ml of dichloromethane was added to the mixture. The organic solution was then washed with an aqueous saturated $NaHCO_3$ solution and brine, sequentially. The organic solution was dried with anhydrous sodium sulfate. After removal of the solvent, the crude product was purified by column chromatography using 20% methanol and 80% ethyl ether as the mobile phase. The yield of **2a** was 37.1%. The ^{1}H-NMR spectrum (300 MHz,$CDCl_3$) of the purified product was: 2.25 (s, 3H), 2.75 - 2.95 (m, 6H), 3.58 - 3.65 (m, 4H), 3.82 - 3.83 (s, 6H), 6.48 - 6.51 (s, 1H), 6.62 - 6.63 (s, 1H), 6.82 - 6.85 (d, 1H), 7.02 (s, 1H), 7.08 (s, 1H), 7.20 (d, 1H). LCMS *m/e*: 371.2 (M+H).

EXAMPLE 10

[0044] This example illustrates synthesis of 5-Bromo-N-[4-(6,7-dimethoxy-3,4-dihydro-1*H*-isoquinolin-2-yl)-butyl]-2-hydroxy-3-methoxy-benzamide (compound 2b).

[0045] Compound **2b** was prepared from 5-bromo-2-hydroxy-3-methoxy-benzoic acid and **1b** as described above for **2a**. The yield of **2b** was 16.7%. The ^{1}H-NMR spectrum (300 MHz,$CDCl_3$) of the purified product was: 1.73 - 1.76 (m, 4H), 2.57 - 2.59 (m, 2H), 2.76 - 2.81 (m, 4H), 3.45 - 3.47 (m, 2H), 3.58 - 3.61 (m, 2H), 3.82 (s, 3H), 3.86(s, 3H), 3.88 (s, 3H), 6.48 -6.51 (t, 1H), 6.56 - 6.59(t, 1H), 6.97 - 7.00 (m, 1H), 7.07 - 7.10 (m, 1H). LCMS *m/e*: 493.10 (M+H).

EXAMPLE 11

[0046] This example illustrates synthesis of N-[4-(6, 7-dimethoxy-3,4-dihydro-1*H*-isoquinolin-2-yl)-butyl]-2-hydroxy-5-methyl-benzamide (compound 2c).

[0047] Compound **2c** was prepared from 2-hydroxy-5-methyl-benzoic acid and **1b** as described above for **2a**. The yield of **2c** was 45%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.75 (m, 4H), 2.12 (s, 3H), 2.58 (m, 2H), 2.75 – 2.77 (m, 2H), 2.82 – 2.84 (m, 2H), 3.40 – 3.50 (m, 2H), 3.58 (s, 2H), 3.83 (s, 3H), 3.85 (s, 3H), 6.50 (s, 1H), 6.60 (s, 1H), 6.85 – 6.88 (d, 1H), 7.06 (s, 1H), 7.13 – 7.16 (d, 2H), 7.61 (s, 1H). LCMS *m/e*: 399.20 (M+H).

EXAMPLE 12

[0048] This example illustrates synthesis of N-[4-(6, 7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-hydroxy-5-bromo-benzamide (compound 2d).

[0049] Compound **2d** was prepared form 5-bromo-2-hydroxy-benzoic acid and **1b** as described above for **2a.** The yield of **2d** was 28.0%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.71 – 1.81 (m, 4H), 2.55-2.85 (m, 6H), 3.44 – 3.48 (m, 2H), 3.58 – 3.60 (m, 2H), 3.82 (s, 3H), 3.89 (s, 3H), 6.50 (s, 1H), 6.59 (s, 1H), 6.82 – 6.84 (d, 1H), 7.30–7.40 (d, 1H), 7.52 (d, 1H), 8.30 (s,1H). Anal. ($C_{22}H_{27}BrN_2O_4.1.25H_2O$) C, H, N.

EXAMPLE 13

[0050] This example illustrates synthesis of N-[4-(6, 7-dimethoxy-3, 4-dihydro-1*H*-isoquinolin-2-yl)-butyl]-2-hydroxy-5-iodo-benzamide (2e).

[0051] Compound **2e** was prepared form 2-hydroxy-5-iodo-benzoic acid and **1b** as described above for **2a.** The yield of **2e** was 27.0%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.69 – 1.81 (m, 4H), 2.54 – 2.65 (m, 2H), 2.75 – 2.83 (m, 2H), 3.44 – 3.48 (m, 2H), 3.58 (s, 2H), 3.82 (s, 3H), 3.85 (s, 3H), 6.50 (s, 1H), 6.58 (s, 1H), 6.70 – 6.74 (d, 1H), 7.54 – 7.55 (d, 1H), 7.65 – 7.67 (d, 1H), 8.20 (s, 1H). Anal. ($C_{22}H_{27}IN_2O_4.0.75H_2O$) C, H, N.

EXAMPLE 14

[0052] This example describes a general method for synthesis of the substituted 2-(2-fluoroethoxy) benzoic acid amides, 3a-e, in particular compound 3a.

[0053] To synthesize [N-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-ethyl]-2-(2-fluoro-ethoxy)-5-methyl-benzamide (compound 3a), potassium carbonate (792.5 mg, 4.88 mmol) was added to a solution of **2a** (278 mg, 0.75 mmol) and 2-bromo-1-fluoroethane (620 mg, 4.88 mmol) in acetone (60 mL). The reaction mixture was refluxed for 48 h until the reaction was complete as determined by thin layer chromatography with 5% methanol and 95% ethyl ether as the mobile phase. The solvent was evaporatored, 30 ml of water added to the flask, and then the mixture extracted with dichloromethane (25 mL x 3). After the organic layer was dried with anhydrous sodium sulfate, the crude product was purified by column chromatography using 5% methanol and 95% ethyl ether as the mobile phase. The yield of **3a** was 90%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 2.33 (s, 3H), 2.64 - 2.80 (m, 6H), 3.63 - 3.75 (m, 4H), 3.84 (s, 3H), 3.86 (s, 3H), 4.16 (m, 1H), 4.21 (m, 1H), 4.41 (m, 1H), 4.60 (m, 1H), 6.55 (s, 1H), 6.61 (s, 1H), 6.78 - 6.81 (d, 1H), 7.20 (d, 1H), 8.00 (s, 1H), 8.28 (s, 1H). LCMS *m/e*: 417.22 (M+H). For the in vitro binding experiments, the free base was converted into the hydrochloride salt; m.p. 159-161 °C, Anal. ($C_{23}H_{30}ClFN_2O_4$). C. H. N.

EXAMPLE 15

[0054] This example illustrates synthesis of **5-**bromo-N-[4-(6,7-dimethoxy-3,4-dihydro-1*H*-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-3-methoxy-benzamide (compound 3b).

[0055] Compound **3b** was prepared from **2b** as described for **3a** above. The yield was 50%. The ^{1}H-NMR spectrum (300 MHz,$CDCl_3$) of the purified product was: 1.64(m, 4H), 2.48-2.52(t, 3H), 2.64-2.66(t, 3H), 2.74-2.78(t, 3H), 3.42-3.55(m, 4H), 3.79-3.87(m, 9H), 4.19-4.22(t,2H), 4.29-4.32(t, 2H), 4.58-4.61(t, 2H), 4.74-4.77(t, 2H), 6.47(s, 1H), 6.54(s, 1H), 7.07(d, 1H), 7.78(d, 1H), 8.10(s, 1H). For the in vitro binding experiments, the free base was converted into the oxalic acid salt; m.p. 127-129 °C. LCMS *m/e*: 590.30 (M+Li). Anal. ($C_{26}H_{33}BrFN_2O_7$).

EXAMPLE 16

[0056] This example illustrates synthesis of N-[6,7-dimethoxy-3,4-dihydro-1*H*-isoquinolin-2-yl-butyl]-2-(2-fluoro-ethoxy)-5-methyl-benzamide (compound 3c).

[0057] Compound **3c** was prepared from **2c** as described above for **3a**. The yield of **3c** was 67%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.67 - 2.00 (m, 4H), 2.33 (s, 3H), 2.51 - 2.56 (t, 3H), 2.67 - 2.72 (t, 3H), 3H), 2.78 - 2.82 (t, 3H), 3.48 - 3.54 (m, 4H), 3.82 (s, 3H), 3,83 (s, 3H), 4.21 - 4.24 (t, 1H), 4.30 - 4.33 (t, 1H), 4.68 - 4.71 (t, 1H), 4.84 - 4.87 (t, 1H), 6.49 (s, 1H), 6.57 (s, 1H), 6.79 - 6.82 (d, 1H), 7.20 (m, 1H), 7.96 (s, 1H), 7.99 - 7.80 (d, 1H). LCMS *m/e*: 445.25 (M+H). For the in vitro binding experiments, the free base was converted into the oxalic acid salt; m.p. 131 - 133 °C. Anal. ($C_{26}H_{34}FN_2O_6$).

EXAMPLE 17

[0058] This example illustrates synthesis of N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-5-bromo-benzamide (compound 3d).

[0059] Compound **3d** was prepared from **2d** as described above for **3a**. The yield of **3d** was 38.90%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.57 – 1.80 (m, 4H), 2.62 - 2.66 (m, 3H), 2.78 - 2.82 (m, 3H), 3.48 - 3.51 (m, 2H), 3.60 – 3.64 (m, 2H), 3.82 (s, 3H), 3,83 (s, 3H), 4.21 - 4.25 (t, 1H), 4.31 - 4.35 (t, 1H), 4.70 - 4.74 (t, 1H), 4.86 - 4.90 (t, 1H), 6.49 (s, 1H), 6.57 (s, 1H), 6.78 - 6.82 (d, 1H), 7.48 – 7.52 (d, 1H), 7.96 (s, 1H), 8.26 (d, 1H). LCMS *m/e*: 509.1 (M+H). For the in vitro binding experiments, the free base was converted into the oxalic acid salt; m.p. 119 - 121 °C. Anal. ($C_{25}H_{31}BrFN_2O_6$).

EXAMPLE 18

[0060] This example illustrates synthesis of N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-5-iodo-benzamide (compound 3e).

[0061] Compound **3e** was prepared from **2e** as described above for **3a**. The yield of **3e** was 41.4%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.61 – 1.72 (m, 4H), 2.46 – 2.52 (m, 2H), 2.67 – 2.71 (m, 2H), 2.76 – 2.78 (m, 2H), 3.41 – 3.47 (m, 2H), 3.52 (t, 2H), 3.80 (s, 3H), 3.83 (s, 3H), 4.19 – 4.21 (m, 1H), 4.27 – 4.31 (m, 1H), 4.67 – 4.71 (m, 1H), 4.83 – 4.86 (m, 1H), 6.46 (s, 1H), 6.55 (s, 1H), 6.62 – 6.66 (d, 1H),7.62 – 7.67 (m, 1H), 7.87 (s, 1H), 8.40 – 8.41 (d, 1H). LCMS *m/e*: 557.13 (M+H). For the in vitro binding experiments, the free base was converted into the oxalic acid salt; m.p. 121 - 123 °C. Anal. ($C_{25}H_{31}FIN_2O_6$).

EXAMPLE 19

[0062] This example describes a general method for synthesis of the substituted 5-bromo-benzoic acid derivatives into their substituted 5-tributylstannanyl benzoic acid derivatives**.** in particular compound 3g.

[0063] To synthesize N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-3-methoxy-5-tributylstannanyl-benzamide (compound 3g), Nitrogen was bubbled for 5–10 min through a solution of **3b** (200mg, 0.371 mmol) in 20 ml fresh distilled toluene. The whole system was covered with aluminum foil. Tetrakis(triphenylphosphine palladium(0) [$(PPh_3)_4Pd(0)$] (42 mg, 0.036 mmol) and bis(tributytin) $[Sn(C_4H_9)_3]_2$ (575 mg, 0.99 mmol) was added to the reaction mixture and heated overnight at 110^{0}C with an oil bath. Thin layer chromatography with 45% hexane, 45% ethyl ether and 10% methanol as the mobile phase was used to assess when the reaction was complete. After quenching the reaction, the crude product was purified on a silica gel column to isolate the tin intermediate, **3g**. The yield of **3g** was 64%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 0.87 – 1.58 (m, 27H), 1.61 – 1.69 (m, 4H), 2.56 (s, 2H), 2.72 (s, 2H), 2.81 – 2.82 (s, 2H), 3.47 – 3.49 (d, 2H), 3.57 (s, 2H), 3.83 (s, 6H), 3.88 (s, 3H), 4.25 (d, 1H), 4.37 (s, 1H), 4.62 – 4.65(s, 1H), 4.78 – 4.81 (s, 1H), 6.51 (s, 1H), 6.58 (s, 1H), 7.81 (s, 1H), 8.07 (s, 1H). LCMS *m/e*: 747.60 (M-H).

EXAMPLE 20

[0064] This example illustrates a general method for converting the tin precursor of the benzoic acid derivatives into their corresponding iodine substituted benzoic acid derivatives, in particular compound 3f.

[0065] To synthesize (N-[4-(6,7-Dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-3-methoxy-5-iodo-benzamide (3f), **a** solution of iodine in $CHCl_3$ (5 mL, 0.5 M) was added dropwise to a solution of the tin precursor, **3g** (258 mg, 0.34 mmol) in 20 ml CH_2Cl_2 until the color of the solution persisted. The reaction was stirred at room temperature for 30 min, and a solution of 5% aqueous $NaHSO_3$ was added until the solution was colorless. The mixture was extracted with CH_2Cl_2, and the organic layers washed with brine before being dried with Na_2SO_4. The organic layers were then concentrated under vacuum and purified using a silica gel column with 15 % methanol and 85 % ether as the mobile phase to isolate **3f.** The yield of **3f** was 36%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.60 – 1.80 (m, 2H), 1.80 – 2.10 (m, 4H), 3.19 – 3.2 (m, 2H), 3.40 – 3.50 (m, 2H), 3.68 (m, 2H), 3.83 (m, 2H), 3.92 (s, 3H), 3.95 (s, 3H), 3.99 (s, 3H), 4.26 (t, 1H), 4.37 (s, 1H), 4.65 (s, 1H), 4.80 (s, 1H), 6.50 (s, 1H), 6.58 (s, 1H), 7.28 (d, 1H), 8.02 (d, 1H), 8.21 (s, 1H). LCMS *m/e*: 587.14 (M+H). For the in vitro binding experiments, the free base was converted into the oxalic acid salt; m.p. 125 - 127 °C. Anal. ($C_{26}H_{33}FIN_2O_7$).

EXAMPLE 21

[0066] This example describes a general method for converting the substituted 2-hydroxy benzoic acid derivatives into their substituted 2-(2-hydroxy-ethoxy)-benzoic acid amides, in particular compound 4c.

[0067] To synthesize N-[4-6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-hydroxy-ethoxy)-5-methyl-benzamide (compound 4c), anhydrous potassium carbonate (546.0 mg, 3.26 mmol) was added to a solution of **2c** (200.0 mg, 0.5 mmol) and 2-bromoethyl acetate (547.0 mg, 3.27 mmol) in 60 mL of acetone. The reaction mixture was refluxed for 48 h under nitrogen. After 48 h, thin layer chromatography with 15% methanol and 85% ether as the mobile phase indicated that the reaction was complete. After evaporating the solvent, the residue was dissolved in 30 ml of water and extracted with ethyl acetate (20 x 3 mL). Then the organic component was washed with brine, dried with anhydrous sodium sulfate, concentrated, and the final product purified on a silica gel column to isolate the 2-{2-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butylcarbamoyl]-4-methyl-phenoxy}-ethyl ester. The yield of this intermediate was 82.2%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.70 (m, 4H), 2.01 (s, 3H), 2.33 (s, 3H), 2.56 (m, 2H), 2.71 – 2.73 (m, 2H), 2.81 (m, 2H), 3.50 – 3.52 (m, 2H), 3.55 (s, 2H), 3.83 (s, 3H), 3.84 (s, 3H), 4.23 (t, 2H), 4.50 (t, 2H), 6.50 (s, 1H), 6.58 (s, 1H), 6.78 -6.82 (d, 1H), 7.18 – 7.25 (d, 1H), 7.95 (s, 1H), 8.02 (s, 1H).

[0068] NaOH (30 mg, 0.75mmol) was added to a solution of this intermediate (182 mg, 0.375 mmol) in 20 mL of methanol and 10 mL of water. The reaction mixture was stirred overnight until the reaction was complete. Then 0.375 mL of 2 N HCl was added to neutralize the solution. After evaporating the solvent, the residue was dissolved in 60 mL of ethyl acetate. The solution was washed first with water, then brine, and finally dried with anhydrous sodium sulfate. After evaporating the solvent, the crude product was purified on a silica gel column. The yield of **4c** was 96%. The ^{1}H-NMR spectrum (300 MHz,$CDCl_3$) of the purified product was: 1.68 - 1.85 (m, 4H), 2.39 (s, 3H), 2.45 (s, 1H), 2.51 - 2.61 (m, 2H), 2.80 - 2.87 (m, 4H), 3.45 - 3.61 (m, 4H), 3.76 - 3.80 (t, 2H), 3.83 (s, 3H), 3.85 (s, 3H), 3.83 (s, 3H), 4.05 – 4.08 (t, 2H), 6.49 (s, 1H), 6.60 (s, 1H), 6.79 – 6.83 (d, 1H), 7.15 – 7.19 (d, 2H), 7.93 (s, 1H), 8.30 (s, 1H).

EXAMPLE 22

[0069] This example illustrates synthesis of 5-Bromo-N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-hydroxy-ethoxy)-benzamide (compound 4d).

[0070] Compound **4d** was prepared from **2d** as described above for **4c**. The yield of compound **4d** was 70%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.65 - 1.90 (m, 4H), 2.66 – 2.70 (m, 2H), 2.92 (m, 4H), 3.51 – 3.54 (m, 2H), 3.72 (m, 2H), 3.72 – 3.81 (t, 2H), 3.83 (s, 3H), 3.85 (s, 3H), 4.05 – 4.09 (t, 2H), 6.51 (s, 1H), 6.60 (s, 1H), 6.77 – 6.81 (d, 1H), 7.44 – 7.48 (d, 2H), 8.17 (s, 1H), 8.30 (s, 1H).

EXAMPLE 23

[0071] This example illustrates synthesis of N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-hydroxy-ethoxy)-5-iodo-benzamide (compound 4e).

[0072] Compound **4e** was prepared from **2e** as described above for **4c**. The yield of **4e** was 77%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.60 – 1.90 (m, 4H), 2.63 – 2.66 (m, 3H), 2.90 (s, 2H), 3.52 – 3.55 (m, 2H), 3.70 (m, 2H), 3.79 – 3.82 (t, 2H), 3.83 (s, 3H), 3.85(s, 3H), 4.08 – 4.10 (t, 2H), 6.50 (s, 1H), 6.60 (s, 1H), 6.67– 6.70 (d, 1H), 7.60–7.70 (d, 1H), 8.30 (s, 1H), 8.40 -8.41 (d, 1H).

EXAMPLE 24

[0073] This example describes a general method for converting the substituted 2-hydroxy-ethoxy benzoic acid amides of the present teachings to their methanesulfonic acid esters, in particular compound 5c.

[0074] To synthesize 2-(2-(4-(6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-yl)butylcarbamoyl)-4-methylphenoxy)ethyl methanesulfonate (compound 5c), methanesulfonic chloride (120 mg, 1.04 mmol) was added to an ice-water cooled solution of **4c** (354 mg, 0.8 mmol) and triethylamine (242 mg, 2.4 mmol) in 30 mL of dichloromethane. The reaction mixture was stirred for 3 h until thin layer chromatography using 5% methanol and 95% dichloromethane as the mobile phase indicated that the reaction was complete. After 3 h, 20 mL of dichloromethane was added, the solution was washed with first a saturated sodium carbonate aqueous solution (20 mL x 3), then brine, and finally dried with anhydrous sodium sulfate. After evaporating the solvent, the crude product was purified on a silica gel column to isolate **5c**. The yield of **5c** was 81.6%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 2.07 – 2.10 (m, 4H), 2.71 (s, 3H), 2.80 – 3.00 (m, 2H), 3.08 – 3.20 (m, 4H), 3.40 (m, 3H), 3.80 – 4.00 (m, 4H), 4.20 (s, 3H), 4.22 (s, 3H), 4.60 – 4.68 (t, 2H), 4.95 – 4.97 (t, 2H), 6.88 (s, 1H), 6.95 (s, 1H), 7.15 – 7.18 (d, 1H), 7.50 – 7.60 (d, 1H), 8.20 (s, 1H), 8.19 (s, 1H). Anal. ($C_{26}H_{36}N_2O_7S$) C, H, N.

EXAMPLE 25

[0075] This example illustrates synthesis of 2-(4-bromo-2-(4-(6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-yl)butylcarbamoyl)phenoxy)ethyl methanesulfonate (compound 5d).

[0076] Compound **5d** was prepared from **4d** as described above for **5c**. The yield of **5d** was 77%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.72 – 1.75 (m, 4H), 2.61 – 2.68 (m, 2H), 2.85 (s, 3H), 3.05 (s, 2H), 3.42 – 3.58 (m, 4H), 3.68 (s, 2H), 3.82 (s, 3H), 3.83 (s, 3H), 4.29(t, 2H), 4.60(t, 2H), 6.50 (s, 1H), 6.57 (s, 1H), 6.70–6.77 (d, 1H), 7.45–7.55 (d, 1H), 7.95 (s, 1H), 8.20-8.22 (d, 1H). LCMS *m/e*: 585.10 (M+H).

EXAMPLE 26

[0077] This example illustrates synthesis of 2-(2-(4-(6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-yl)butylcarbamoyl)-4-iodophenoxy)ethyl methanesulfonate (compound 5e).

[0078] Compound **5e** was prepared from **4e** as described above for **5c**. The yield of **5e** was 80%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 1.60 – 1.80 (m, 4H), 2.52 – 2.55 (m, 2H), 2.67 – 2.70 (m, 2H), 2.70 – 2.78 (m, 2H), 3.02(s, 3H), 3.46 – 3.52 (m, 4H), 3.82 (s, 3H), 3.83 (s, 3H), 4.23 – 4.26 (t, 2H), 4.56 – 4.60 (t, 2H), 6.48 (s, 1H), 6.55 (s, 1H), 6.60 – 6.64 (d, 1H), 7.64 – 7.68 (d, 1H), 7.85 (s, 1H), 8.38-8.39 (d, 1H). LCMS *m/e*: 633.10 (M+H).

EXAMPLE 27

[0079] This example illustrates synthesis of 2-(2-Acetoxy-ethoxy)-5-bromo-3-methoxy-benzoic acid methyl ester (compound 7).

[0080] To prepare this compound, initially 1.0 mL of 98% concentrated sulfuric acid was added to a solution of 5-bromo-2-hydroxy-3-methoxy-benzoic acid, (compound **6)** (1.0 g, 4.0 mmol) in 50 ml of methanol. The reaction mixture was refluxed overnight until thin layer chromatography using 20% ethyl acetate and 80% hexane as the mobile phase indicated that the reaction was complete. After evaporating the methanol, the residue was dissolved in 60 mL of ethyl acetate and washed with a saturated $NaHCO_3$ aqueous solution and then brine. After drying with anhydrous sodium sulfate, the solution was concentrated, and the crude product was purified on a silica gel column to isolate the intermediate, 5-bromo-2-hydroxy-3-methoxy-benzoic acid methyl ester. The yield of this intermediate was 94%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 3.88 (s 3H), 3.95 (s, 3H), 7.09 (d, 1H), 7.55 (t, 1H), 10.96 (d, 1H).

[0081] Potassium carbonate (2.90g, 21.0 mmol) was added to a solution of the above intermediate (0.84 g , 3.23 mmol) and 2-bromoethyl acetate (3.5 g, 20.96 mmol) in 60 mL of acetone. The reaction mixture was refluxed for 72 h until thin layer chromatography using 20% ethyl acetate and 80% hexane as the mobile phase indicated that the reaction was complete. After evaporating the solvent, the residue was dissolved in 30 mL of water and then extracted with ethyl acetate (25 mL x 3). The organic solution was dried with anhydrous sodium sulfate, resuspended, and purified on a silica gel column. The yield of **7** was 78%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 2.10 (s, 3H), 3.86 (s, 3H), 3.88 (s, 3H), 4.21 – 4.24 (t, 2H), 4.38 – 4.41 (t, 3H), 7.14 (s, 1H), 7.45 (s, 1H).

EXAMPLE 28

[0082] This example illustrates synthesis of 2-(2-Acetoxy-ethoxy)-5-iodo-3-methoxy-benzoic acid methyl ester (compound 8).

[0083] To synthesize compound 8, nitrogen was bubbled for 5-10 min through a solution of 2-(2-acetoxy-ethoxy)-5-bromo-3-methoxy-benzoic acid methyl ester, (compound **7)** (270 mg, 0.778 mmol) in 20 mL of freshly distilled toluene. The reaction system was covered with aluminum foil. Tetrakis(triphenylphosphine) palladium(0) [$(PPh_3)_4Pd(0)$] (100 mg, 0.087 mmol) and bis(tributlytin) [$Sn(C_4H_9)_3]_2$ (899mg, 1.55 mmol) were added to the reaction mixture and heated overnight at 110^{0}C in an oil-bath while stirring. After quenching, thin layer chromatography using 15% ethyl acetate and 85% hexane as the mobile phase indicated that the reaction was complete. The product was then purified on a silica gel column to isolate the tin precursor, 2-(2-acetoxy-ethoxy)-3-methoxy-5-tributylstannanyl-benzoic acid methyl ester. The yield of the tin precursor was 37.3%. The ^{1}H-NMR spectrum

(300 MHz, $CDCl_3$) of the purified product was: 0.8 – 1.75 (m, 27H), 2.10 (s, 3H), 3.87 (s, 3H), 3.89 (s, 3H), 4.24 – 4.27 (t, 2H), 4.38 – 4.41 (t, 2H), 7.09 – 7.30 (s 1H), 7.35 (s 1H).

[0084] A solution of iodine in $CHCl_3$ (5 mL, 0.5 M) was added dropwise to a solution of the above tin precursor (680 mg, 1.22 mmol) in 20 mL of CH_2Cl_2 until the color of the solution persisted. Then the reaction was stirred at room temperature for 30 min, and a quench solution of 5% aqueous $NaHSO_3$ was added until the solution became colorless. The mixture was extracted with CH_2Cl_2, and the organic layers washed with brine and dried by Na_2SO_4. The organic layers were then condensed under vacuum and purified using a silica gel column with 15% ethyl acetate and 85% hexane as the mobile phase. The yield of compound **8** was 90%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 2.10 (s, 3H), 3.85 (s, 3H), 3.88 (s, 3H), 4.21 – 4.25 (t, 2H), 4.37 – 4.41 (t, 2H), 7.29 – 7.30 (s 1H), 7.63 (s 1H).

EXAMPLE 29

[0085] This example illustrates synthesis of 2-(2-Hydroxy-ethoxy)-5-iodo-3-methoxy-benzoic acid (compound 9)

[0086] Compound **9** was prepared from compound **8** as described in the general method for converting the substituted 2-hydroxy benzoic acid derivatives into their substituted 2-(2-hydroxy-ethoxy)-benzoic acid amides (Example 21). The yield of compound **9** was 81%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$) of the purified product was: 3.89 (s, 3H), 3.93 – 3.96 (t, 2H), 4.33 – 4.36 (t, 2H), 7.08 (s, 1H), 7.62 (s, 1H).

EXAMPLE 30

[0087] This example illustrates synthesis of N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-hydroxy-ethoxy)-5-iodo-3-methoxy-benzamide (compound 4f)

[0088] Compound **4f** was prepared from **9** and **1b** as described in **the** general method for synthesis of the substituted 2-hydroxybenzoic acid amides (Example 9). The yield of **4f** was 29%. The ^{1}H-NMR spectrum (300 MHz, $CDCl_3$)of the purified product was: 1.72 – 1.75 (m, 4H), 2.56 (m, 2H), 2.75 – 2.77 (m, 2H), 2.81 – 2.83 (m, 2H), 3.49 – 3.51 (m, 2H), 3.55 (s, 2H), 3.56 – 3.60 (t, 2H), 3.82 (s, 3H), 3.83 (s, 3H), 3.85 (s, 3H), 4.06 – 4.10 (t, 2H), 6.47 (s, 1H), 6.57 (s, 1H), 6.90 (s, 1H), 7.57 (s, 1H), 7.70–7.80 (s, 1H).

EXAMPLE 31

[0089] This example illustrates synthesis of 2-(2-(4-(6,7-dimethoxy-3,4-dihydroisoquinolin-2(1H)-yl)butylcarbamoyl)-4-iodo-6-methoxyphenoxy)ethyl methanesulfonate (compound **5f**).

[0090] Compound **5f** was prepared from **4f** as described in the general method for converting the substituted 2-hydroxy-ethoxy benzoic acid amides of the present teachings to their methanesulfonic acid esters (Example 24). The yield of **5f** was 61%. The ^{1}H-NMR spectrum (300 MHz,$CDCl_3$) of the purified product was: 1.72(m, 4H), 2.55(s, 2H), 2.70(d, 2H), 22.76(d, 2H), 3.05(s, 3H), 3.85(m, 9H), 4.26(m, 2H), 4.49(m, 2H), 6.49(s,1H), 6.55(s, 1H), 6.93(d, 1H), 7.51(m, 1H), 8.02(s, 1H) . LCMS *m/e*: 663.20 (M+H). Anal. ($C_{25}H_{32}FIN_2O_5$) C: Calcd, 51.20; found 36.61; C: Calcd, 5.50; found 4.34; N: Calcd, 4.78; found 3.12.

EXAMPLE 32

[0091] This example illustrates production of [^{18}F]Fluoride.

[0092] [^{18}F]Fluoride was produced in our institution by proton irradiation of enriched ^{18}O water (95%) [reaction: $^{18}O(p, n)^{18}F$] using either a JSW BC-16/8 (Japan Steel Works) or a CS-15 cyclotron (Cyclotron Corp).

EXAMPLE 33

[0093] This example illustrates a general method for labeling the substituted 2-(2-fluoroethoxy) benzoic acid amide analogs with ^{18}F, in particular [^{18}F](N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-3-methoxy-5-iodo-benzamide (compound **[^{18}F]3f).**

[0094] For this synthesis, [^{18}F]fluoride (100 -150 mCi) was added to a 10-mL Pyrex screw cap tube containing 5-6 mg of Kryptofix 222 and 0.75 mg of K_2CO_3. Using HPLC grade acetonitrile (3 × 1.0 mL), the water was azeotropically evaporated from this mixture at 110°C under a stream of argon. After all of the water was removed, a solution of the precursor, **5f,** (1.5-2.0 mg) in DMSO (0.2 mL) was added to the reaction vessel containing the ^{18}F/Kryptofix mixture. A 3 mm glass bead was added to the reaction vessel to insure a more homogeneous heat distribution when the sample was irradiated with microwaves, and the vessel capped firmly on a specially designed remotely operated capping station. After vortexing, the reaction mixture was irradiated with microwaves for 30 - 40 sec at medium power (60 Watts) until the thin layer chromatography scanner with a 25% of methanol and 75% dichloromethane mobile phase indicated that the incorporation yield was 40-60%.

[0095] After adding 6 mL of water and shaking, the solution was loaded on a C-18 reverse phase Waters Oasis cartridge (HLB-6cc) that had previously been rinsed with a solution of 5% methanol in water (5-8 mL). The sample was then rinsed 3 times with 6 mL water to eliminate the unreacted fluoride. The retained activity was eluted with 5-8 mL of acetonitrile. After evaporating the acetonitrile to a volume of <0.5 mL, the sample was loaded on a C-18 Alltech econosil semi-preparative HPLC column (250 x 10 um). The product was eluted with 29% acetonitrile and 71% 0.1M ammonium formate buffer at a flow rate of 4.5 mL/min. The retention time of the **[^{18}F]3f** was ~33 min. The solution containing the **[^{18}F]3f** was concentrated, resuspended in saline, and a 100 uL aliquot sent for quality control analysis before using it in the biodistribution and imaging studies. The entire procedure required ~2 h.

[0096] Quality control analysis was performed on an analytical HPLC system that consisted of an Alltech econosil reversed phase C-18 column (250 x 4.6 mm) with a mobile phase of 35% acetonitrile and 65% 0.1 M ammonium formate buffer at pH 4.0–4.5. At a flow rate of 1.2 mL/min, the **[^{18}F]3f** eluted at 13.2 min with a radiochemical purity of >99%. The labeling yield was ~30% (decay corrected), and the specific activity was >2000 Ci/mmol.

EXAMPLE 34

[0097] This example illustrates synthesis of **[^{18}F]**(N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-5-methyl-benzamide (compound **[^{18}F]**3c)**.**

[0098] Compound **[^{18}F]3c** was prepared from **5c** as described above for **[^{18}F]3f** with the following exceptions. The semi-preparative HPLC mobile phase was 39% methanol and 61% 0.1 M formate buffer. At a flow rate of 3.5 mL/min, the **[^{18}F]3c** eluted at ~33 min with a radiochemical purity of >99%. The labeling yield was ~35% (decay corrected), and the specific activity was >1500 Ci/mmol. The entire procedure took ~2 h.

[0099]To check that the chemical characteristics of **[^{18}F]3c** were identical to the cold standard, **3c,** both compounds were run on the analytical HPLC system with a mobile phase of 52% methanol and 48% 0.1 M formate buffer. At a flow rate of 1.5 mL/min, the two compounds co-eluted with a retention time of 4.7 min.

EXAMPLE 35

[0100] This example illustrates synthesis of [^{18}F](N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-5-bromo-benzamide (compound **[^{18}F]3d).**

[0101] Compound **[^{18}F]3d** was prepared from **5d** as described above for **[^{18}F]3f** with the following exceptions. The semi-preparative HPLC mobile phase was 13% of THF and 87% 0.1 M formate buffer. At a flow rate of 3.5 mL/min, the **[^{18}F]3d** eluted at ~20 min with a radiochemical purity of >98%. The labeling yield was ~30% (decay corrected), and the specific activity was >1500 Ci/mmol. The entire procedure took ~2 h.

[0102] To check that the chemical characteristics of **[^{18}F]3d** were identical to the cold standard, **3d,** both compounds were run on the analytical HPLC system with a mobile phase of 38% acetonitile and 62% 0.1 M formate buffer. At a flow rate of 1.5 mL/min, the two compounds co-eluted with a retention time of ~8 min.

EXAMPLE 36

[0103] This example illustrates synthesis of **[^{18}F]**(N-[4-(6,7-dimethoxy-3,4-dihydro-1H-isoquinolin-2-yl)-butyl]-2-(2-fluoro-ethoxy)-5-Iodo-benzamide (compound **[^{18}F]3e**).

[0104] Compound **[^{18}F]3e** was prepared from **5e** as described above for **[^{18}F]3f** with the following exceptions. The semi-preparative HPLC mobile phase was 15% THF and 85% 0.1 M formate buffer. At a flow rate of 6.0 mL/min, the **[^{18}F]3e** eluted at ~35 min with a radiochemical purity of >99%. The labeling yield was ~30% (decay corrected), and the specific activity was >1500 Ci/mmol. The entire procedure took ~2 h.

[0105] To check that the chemical characteristics of **[^{18}F]3e** were identical to the cold standard, **3e,** both compounds were run on the analytical HPLC system with a mobile phase of 13% acetonitrile and 87% 0.1 M formate buffer. At a flow rate of 2.0 mL/min, the two compounds co-eluted with a retention time of 15.2 min.

EXAMPLE 37

[0106] This example provides an elemental analysis of various compounds of the present teachings. The data are presented in Table V.

Table V. Elemental Analysis

Compounds	Formula	Calculated				Measured		
		C	H	N		C	H	N
2d	$C_{22}H_{27}BrN_2O_4.1.25H_2O$	54.38	6.12	5.77		54.44	5.74	5.69
2e	$C_{22}H_{27}IN_2O_4.0.75H_2O$	50.44;	5.48	5.35		50.45	5.27	5.24
5c	$C_{26}H_{36}N_2O_7S$	59.98	6.97	5.38		59.89	6.94	5.49
3a	$C_{23}H_{30}ClFN_2O_4.H_2O$	58.66	6.85	5.95		58.94	6.65	6.01
3b	$C_{26}H_{33}BrFN_2O_7.0.5H_2O$	52.62	5.77	4.72		53.29	6.27	3.98
3c	$C_{26}H_{34}FN_2O_6.H_2O$	61.52	7.15	5.52		61.84	6.95	5.27
3d	$C_{25}H_{31}BrFN_2O_6.1.5H_2O$	51.64	5.89	4.82		51.32	5.47	4.59
3e	$C_{25}H_{31}FIN_2O_6.1.75H_2O$	47.44	5.49	4.43		47.07	5.09	4.11

EXAMPLE 38

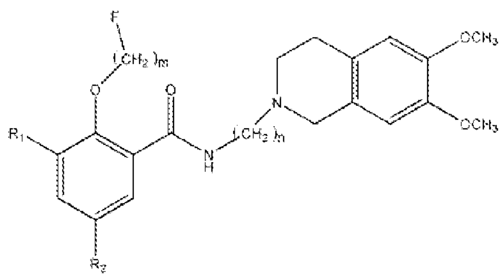
[0107] This example illustrates, in Fig. 9, imaging of a glioma using **[^{18}F]3f** of the present teachings compared to [^{18}F]FDG. Note greater contrast using compound **[^{18}F]3f** as a radiotracer.

Patents and publications discussed herein are hereby incorporated by reference.

CLAIMS

What is claimed is:

1. A fluoroalkoxybenzamide compound of structure



wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof**.**

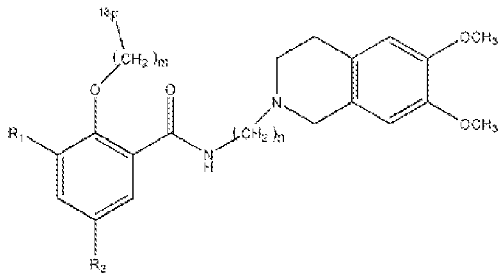
2. A fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 1, wherein m=2, n=4, R_1 is H, and R_2 is CH_3.

3. A fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 1, wherein m=2, n=4, R_1 is H, and R_2 is Br.

4. A fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 1, wherein m=2, n=4, R_1 is H, and R_2 is I.

5. A fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 1, wherein m=2, n=4, R_1 is OCH_3, and R_2 is I.

6. A radiolabelled fluoroalkoxybenzamide compound of structure



wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof.

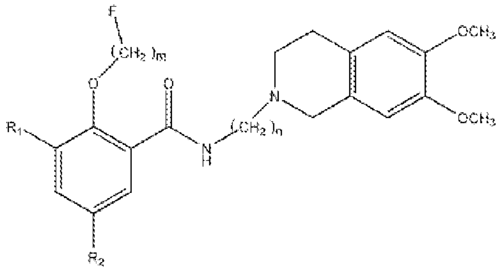
7. A radiolabelled fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 6, wherein m=2, n=4, R_1 is H, and R_2 is CH_3.

8. A radiolabelled fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 6, wherein m=2, n=4, R_1 is H, and R_2 is Br.

9. A radiolabelled fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 6, wherein m=2, n=4, R_1 is H, and R_2 is I.

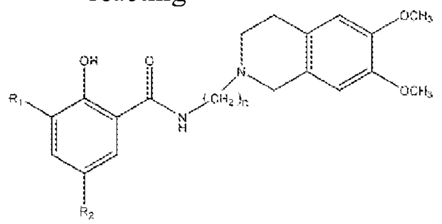
10. A radiolabelled fluoroalkoxybenzamide compound or a salt thereof in accordance with claim 6, wherein m=2, n=4, R_1 is OCH_3, and R_2 is I.

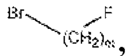
11. A method for synthesizing a fluoroalkoxybenzamide compound of structure

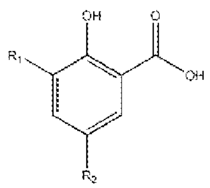
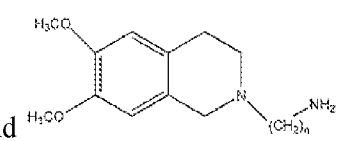
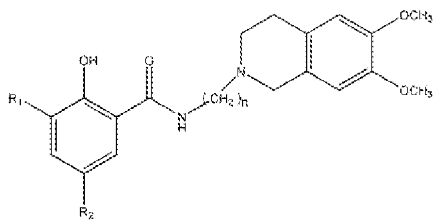


, the method comprising:

reacting



with , wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of Br, Cl and F, **a** C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2**.**

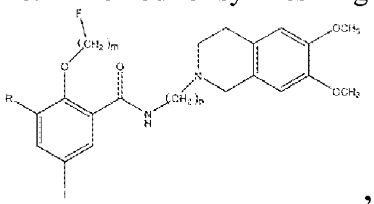
12. A method for synthesizing a fluoroalkoxybenzamide compound in accordance with claim 11, further comprising contacting  and , thereby forming .

13. A method for synthesizing a fluoroalkoxybenzamide compound in accordance with claim 11, wherein m=2, n=2, R_1 is H, and R_2 is CH_3.

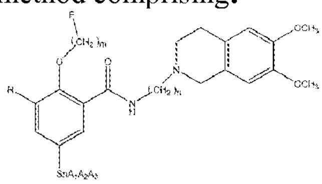
14. A method for synthesizing a fluoroalkoxybenzamide compound in accordance with claim 11, wherein m=2, n=4, R_1 is selected from the group consisting of OCH_3 and H, and R_2 is selected from the group consisting of Br, CH_3, and I.

15. A method for synthesizing a fluoroalkoxybenzamide compound in accordance with claim 11, wherein m=2, n=4, R_1 is H, and R_2 is I.

16. A method for synthesizing a compound of structure



, the method comprising:

iodinating a compound of structure 

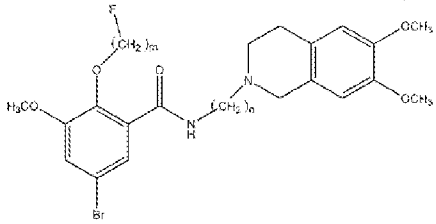
, wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, A_1, A_2 and A_3 are each independently a C_{1-4} alkyl, and R is selected from the group consisting of H and C_{1-4} alkoxy.

17. A method in accordance with of claim 16, wherein m=2 and n=4.

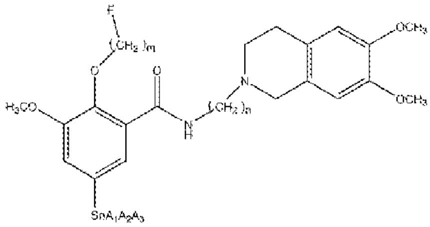
18. A method in accordance with of claim 16, wherein at least one of A_1, A_2 and A_3 is a butyl moiety selected from the group consisting of an n-butyl moiety, an iso-butyl moiety, a sec-butyl moiety, and a tert-butyl moiety.

19. A method in accordance with claim 16, wherein A_1, A_2 and A_3 are each an n-butyl moiety, m=2, n=4 and R is a methoxy.

20. A method in accordance with claim 16, further comprising stannylating a compound **of** structure



to form the compound of structure



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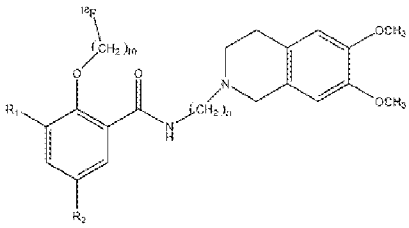
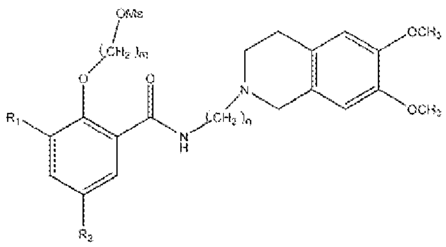
21. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound of structure



, the method comprising**:**

forming a mixture comprising i) an organic solvent, ii) a compound of structure



, iii) $^{18}F^{-}$, iv) 4,7,13,16,21,24-Hexaoxa-1,10-diazabicyclo[8.8.8]-hexacosane and v) a potassium salt, wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2.

22. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound in accordance with claim 21, wherein m=2, n=4, R_1 is selected from the group consisting of H and OCH_3, and R_2 is selected from the group consisting of CH_3, Br and I.

23. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound in accordance with claim 21, wherein m=2, n=4, R_1 is OCH_3, and R_2 is I.

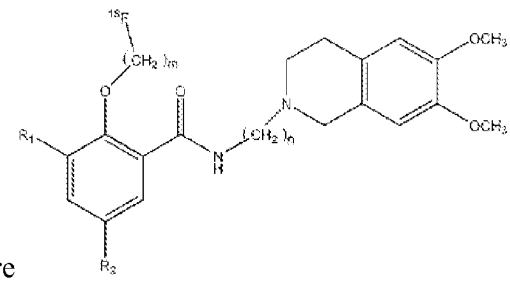
24. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound in accordance with claim 21, wherein the potassium salt is K_2CO_3.

25. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound in accordance with claim 21, wherein the organic solvent is dimethyl sulfoxide.

26. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound in accordance with claim 21, wherein the organic solvent is acetonitrile.

27. A method for synthesizing a radiolabelled fluoroalkoxybenzamide compound in accordance with claim 21, further comprising heating the mixture.

28. A method of imaging a tumor in a mammal, the method comprising:

administering to the mammal a radiolabelled fluoroalkoxybenzamide compound of structure 

, wherein m is an integer from 1 to about

10, n is an integer from 1 to about 10, and R_1 and R_2 are each independently selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof; and

subjecting the mammal to positron emission tomography (PET) scanning.

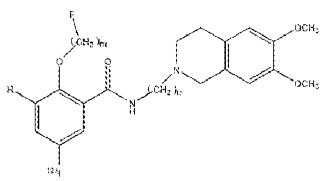
29. A method of imaging a tumor in accordance with claim 28, wherein m=2, n=4, R_1 is selected from the group consisting of H and OCH_3, and R_2 is selected from the group consisting of CH_3, Br and I.

30. A method of imaging a tumor in accordance with claim 28, wherein m=2, n=4, R_1 is H, and R_2 is CH_3.

31. A method of imaging a tumor in accordance with claim 28, wherein m=2, n=4, R_1 is H, and R_2 is Br.

32. A method of imaging a tumor in accordance with claim 28, wherein m=2, n=4, R_1 is H, and R_2 is I.

33. A method of imaging a tumor in accordance with claim 28, wherein m=2, n=4, R_1 is OCH_3, and R_2 is I.

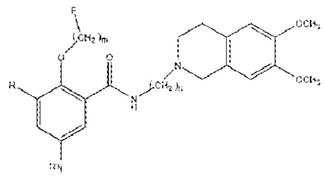
34. A compound of structure  wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R is selected from the group consisting of H,

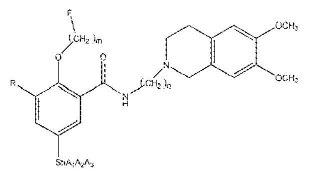
a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof.

35. A compound or salt thereof in accordance with of claim 34, wherein m=2 and n=4.

36. A compound or salt thereof in accordance with claim 34, wherein R is selected from the group consisting of H and C_{1-4} alkoxy.

37. A compound or salt thereof in accordance with claim 34, wherein the C_{1-4} alkoxy is a methoxy.

38. A method for synthesizing a compound of structure , the method comprising:

reacting a compound of structure 
with [^{123}I]NaI and an oxidant, wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, A_1, A_2 and A_3 are each independently a C_{1-4} alkyl, and R is selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2.

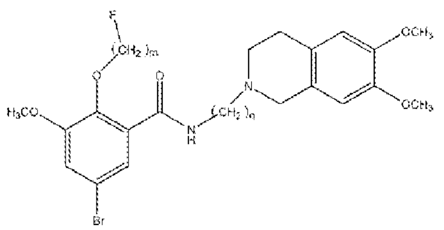
39. A method in accordance with of claim 38, wherein m=2 and n=4.

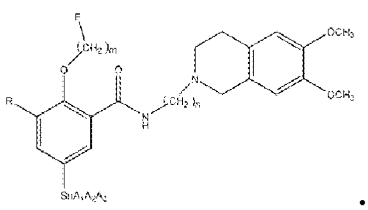
40. A method in accordance with claim 38, wherein R is selected from the group consisting of H and C_{1-4} alkoxy.

41. A method in accordance with claim 40, wherein the C1-4 alkoxy is a methoxy.

42. A method in accordance with of claim 38, wherein at least one of A_1, A_2 and A_3 is a butyl moiety selected from the group consisting of an n-butyl moiety, an iso-butyl moiety, a sec-butyl moiety, and a tert-butyl moiety.

43. A method in accordance with claim 38, wherein A_1, A_2 and A_3 are each an n-butyl moiety, m=2 and n=4

44. A method in accordance with claim 38, further comprising stannylating a compound of structure  to form the compound of structure



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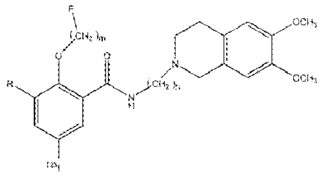
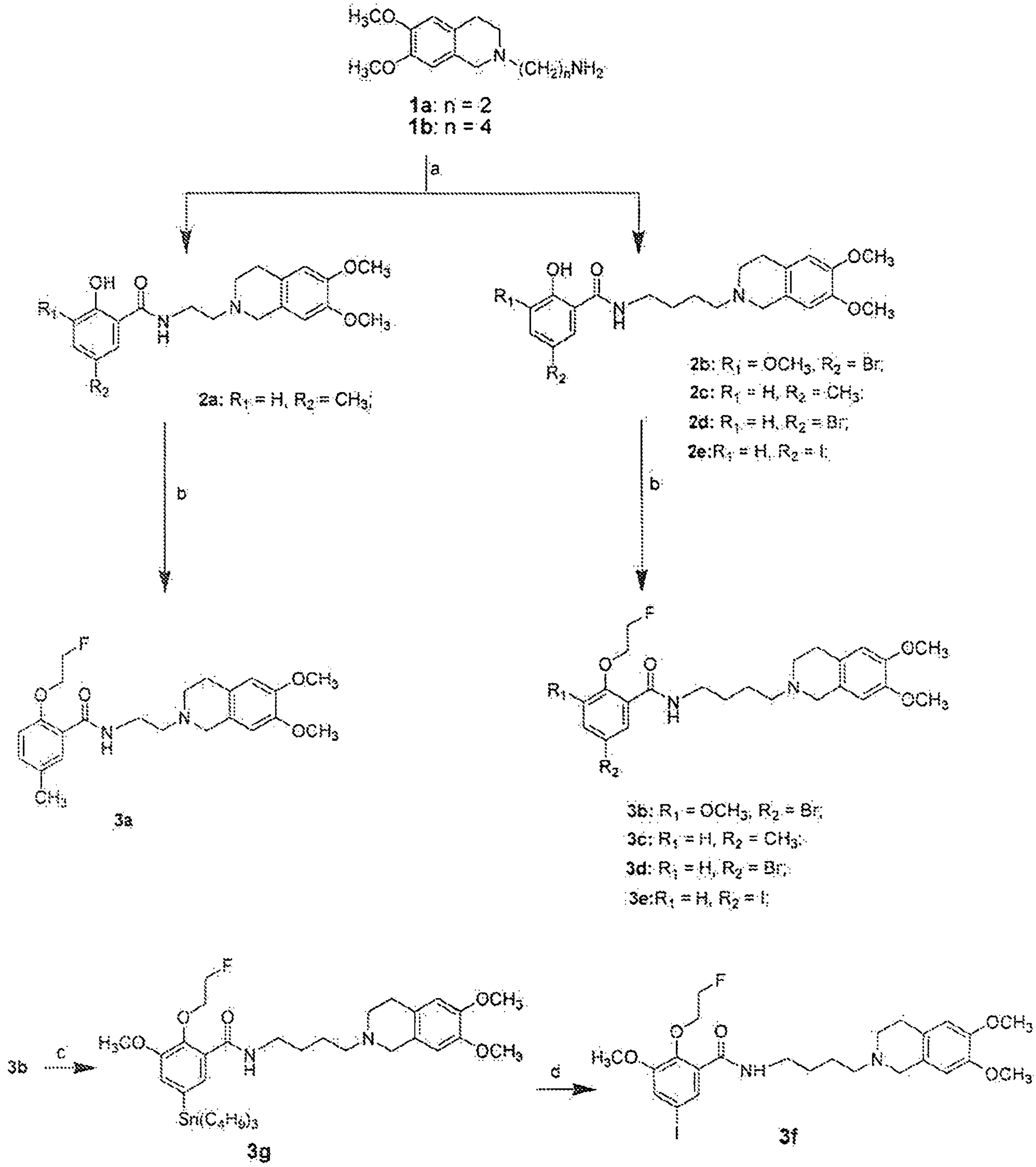
45. A method in accordance with claim 38 wherein the oxidant is selected from the group consisting of peracetic acid, hydrogen peroxide, chloramine T (*N*-Chloro-*p*-toluenesulfonamide sodium salt) and a combination thereof.

46. A method of imaging a solid tumor in a mammal, the method comprising:

administering to the mammal a radiolabelled fluoroalkoxybenzamide compound of structure  wherein m is an integer from 1 to about 10, n is an integer from 1 to about 10, and R is selected from the group consisting of H, a halogen selected from the group consisting of I, Br, Cl and F, a C_{1-4} alkoxy, a C_{1-4} alkyl, a C_{1-4} fluoroalkyl, a C_{1-4} fluoroalkoxy, CF_3, OCF_3, SCH_3, SCF_3, and NH_2, or a salt thereof; and

subjecting the mammal to single photon emission computed tomography (SPECT) imaging.

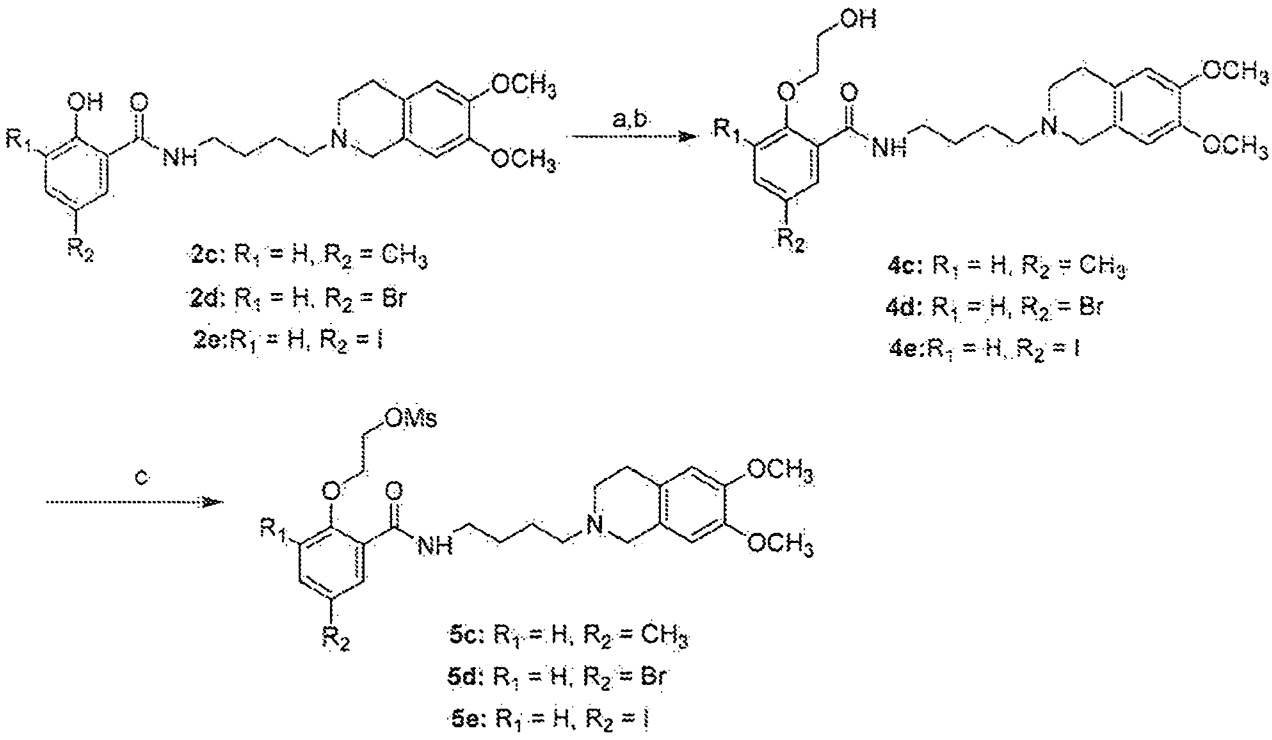
47. A method in accordance with claim 46, wherein the m=2, n=4 and R is selected from the group consisting of H and methoxy.



Reagents: (a) RCOOH, BOP/CH_2Cl_2 or DCC / CH_2Cl_2, rt, 18 hrs; (b) $BrCH_2CH_2F$, K_2CO_3/acetone reflux, over 48hrs; (c)$[Sn(C_4H_9)_3]_2$, $Pd(PPh_3)_4(0)$ / toluene at 110^{0}C; (d) I_2 / CH_2Cl_2, room temperature;

Scheme 1

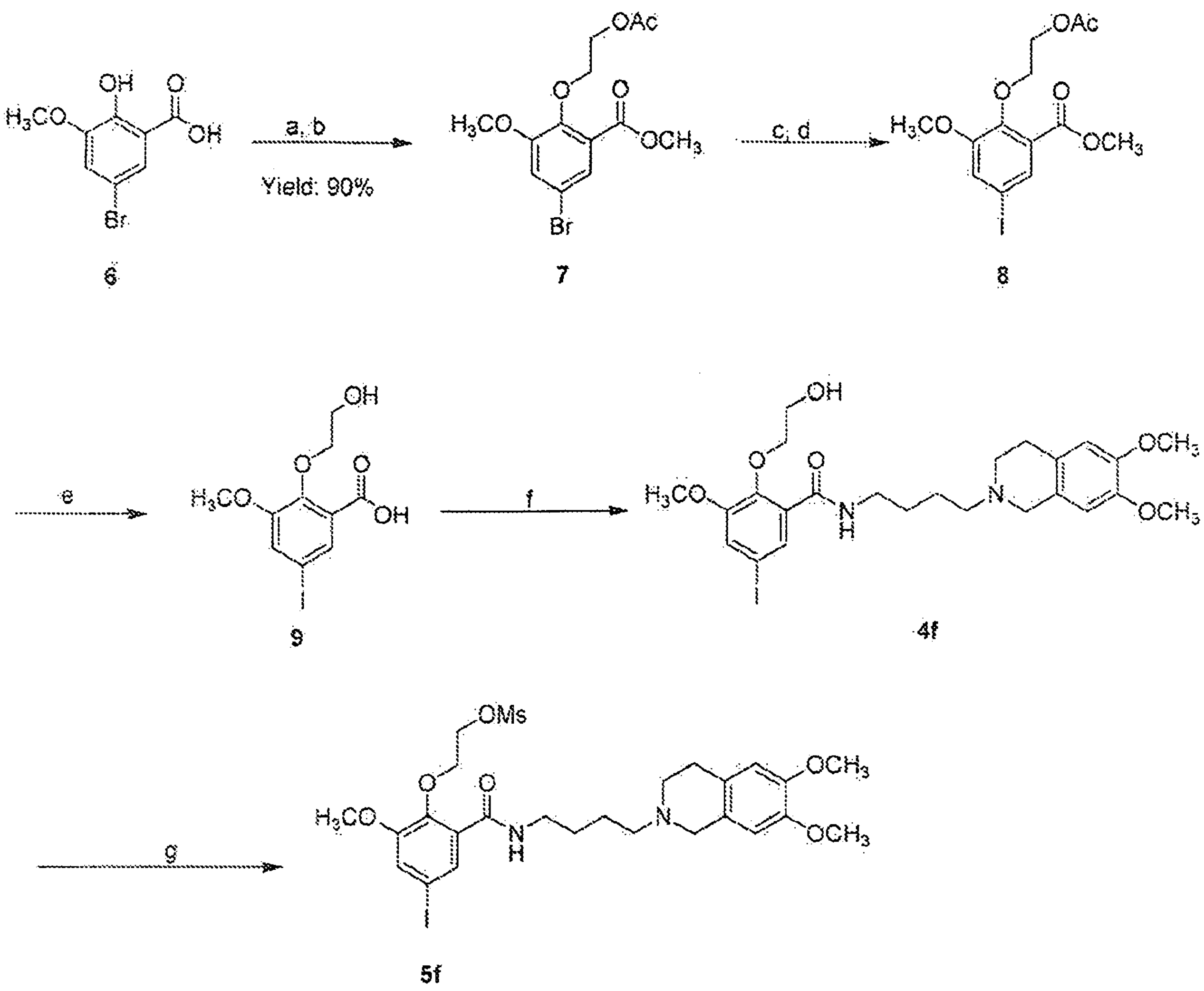
Fig. 1



Reagent: (a) $BrCH_2CH_2OAc$/acetone, K_2CO_3, reflux 18 hrs; (b) NaOH / H_2O, CH_3OH, room temperature; (c) methanesulfonyl chloride, CH_2Cl_2, triethylamine, room temperature.

Scheme II.

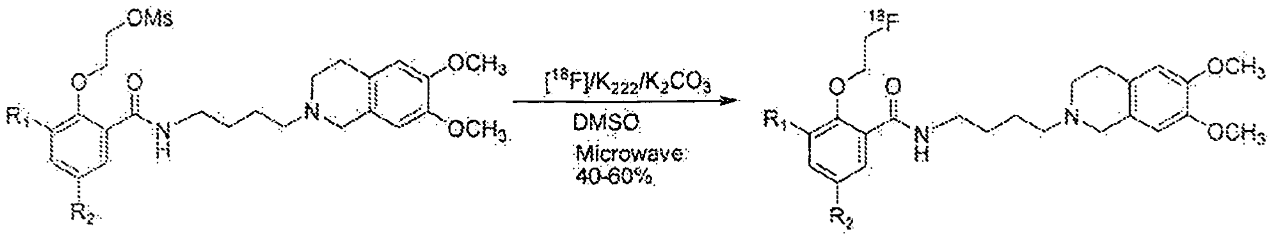
Fig. 2



Reagents: (a) Methanol / 98% H_2SO_4 reflux 18 hrs; (b) $BrCH_2CH_2OAc$/acetone, K_2CO_3, reflux 18 hrs; (c) $[Sn(C_4H_9)_3]_2$, $Pd(PPh_3)_4(0)$ / toluene at 110°C; (d) I_2 / CH_2Cl_2, room temperature; (e) NaOH / H_2O, CH_3OH, room tempearture; (f) **1b**, BOP/CH_2Cl_2 or DCC / CH_2Cl_2, room temperature, 18 hrs; (g) methanesulfonyl chloride; CH_2Cl_2, triethylamine, room temperature, 18hrs.

Scheme III.

Fig. 3

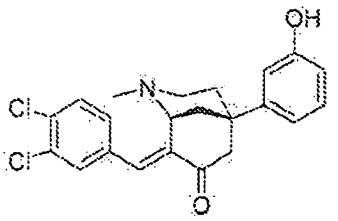


5c: R_1 = H, R_2 = CH_3
5d: R_1 = H, R_2 = Br
5e: R_1 = H, R_2 = I
5f: R_1 = OCH_3, R_2 = I

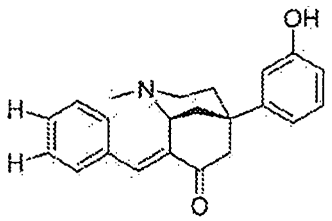
[^{18}F]3c: R_1 = H, R_2 = CH_3
[^{18}F]3d: R_1 = H, R_2 = Br
[^{18}F]3e: R_1 = H, R_2 = I
[^{18}F]3f: R_1 = OCH_3, R_2 = I

Scheme IV.

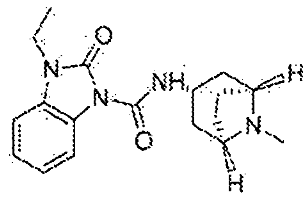
Fig. 4



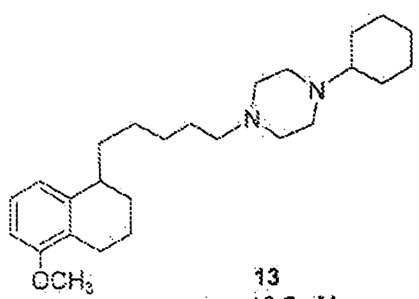
10
σ_1 = 7436 ± 308 nM
σ_2 = 13.4 ± 2.0 nM
$\sigma_1 : \sigma_2$ ratio = 554
LogP = 3.84 (pH = 7.4)



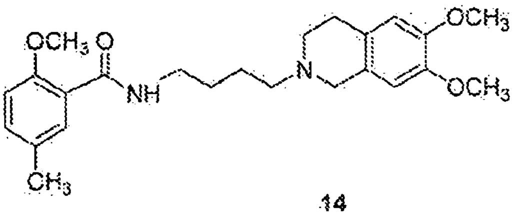
11
σ_1 = 3,063 ± 78 nM
σ_2 = 16.5 ± 2.7 nM
$\sigma_1 : \sigma_2$ ratio = 185
LogP = 3.84 (pH = 7.4)



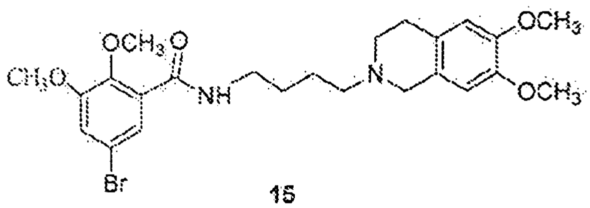
12
σ_1 = 6,300 nM
σ_2 = 32 nM
$\sigma_1 : \sigma_2$ ratio = 200
LogP = -0.96 (pH = 7.4)



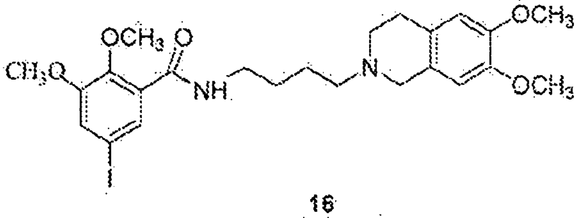
13
σ_1 = 13.6 nM
σ_2 = 0.34 nM
$\sigma_1 : \sigma_2$ ratio = 40
Log P = 5.59 (pH = 7.4)



14
σ_1 = 3078 ± 87 nM
σ_2 = 10.3 ± 1.5 nM
$\sigma_1 : \sigma_2$ ratio = 299
Log P = 2.83 (pH = 7.4)

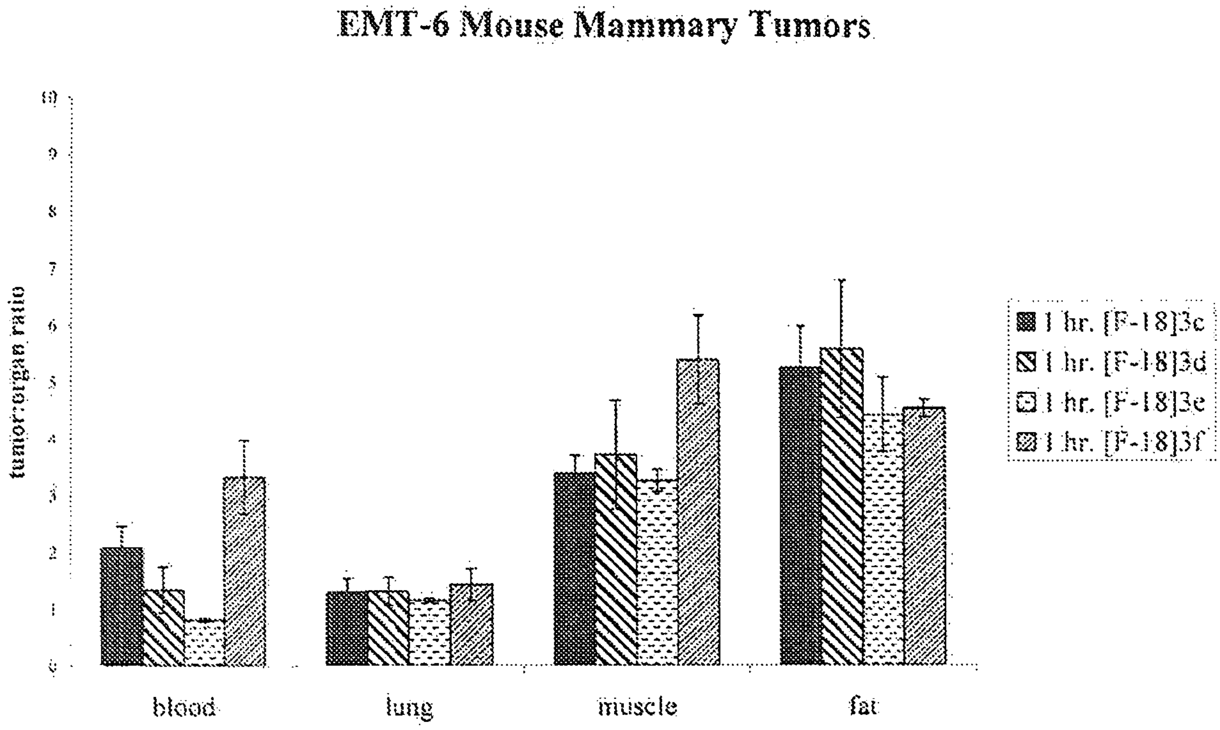


15
σ_1 = 12,900 ± 111 nM
σ_2 = 8.2 ± 1.4 nM
$\sigma_1 : \sigma_2$ ratio = 1575
Log P = 3.3 (pH = 7.4)



16
σ_1 = 554 nM
σ_2 = 11 nM
$\sigma_1 : \sigma_2$ ratio = 50
Log P = 3.94 (pH = 7.4)

Fig. 5


EMT-6 Mouse Mammary Tumors
tumor:organ ratio
0
1
2
3
4
5
6
7
8
9
10
blood
lung
muscle
fat
1 hr. [F-18]3c
1 hr. [F-18]3d
1 hr. [F-18]3e
1 hr. [F-18]3f

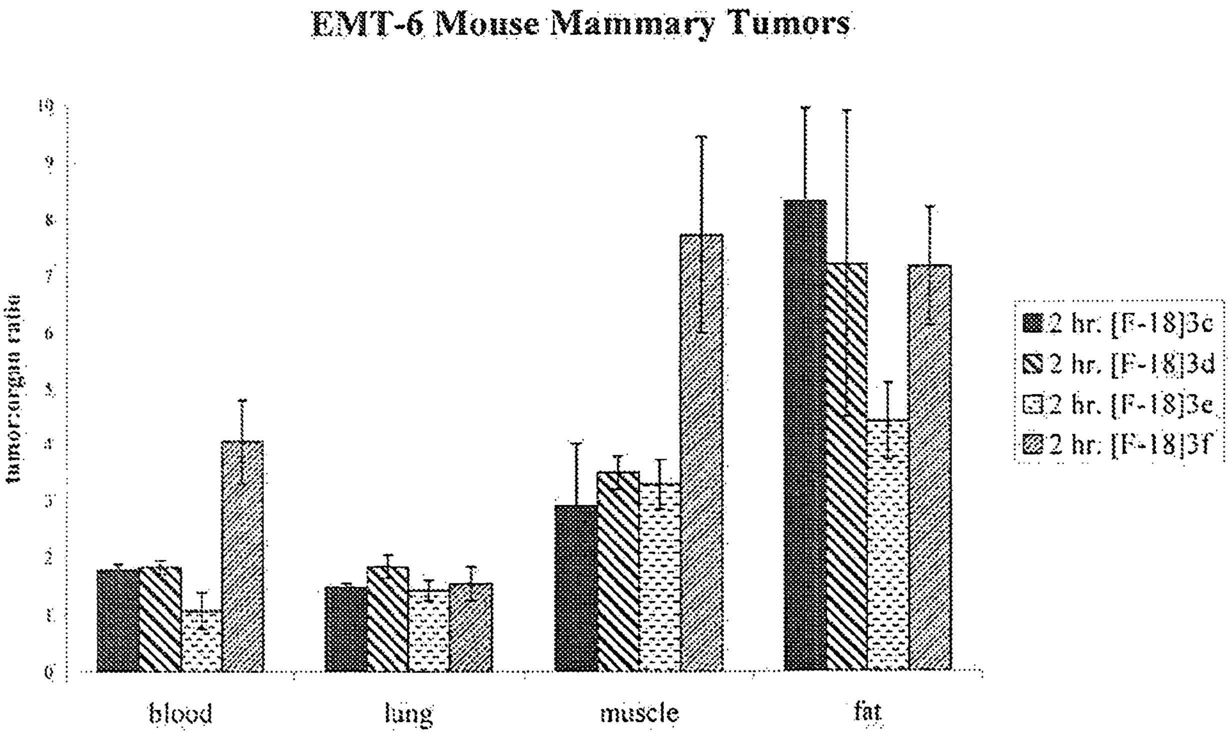

EMT-6 Mouse Mammary Tumors
tumor:organ ratio
0
1
2
3
4
5
6
7
8
9
10
blood
lung
muscle
fat
2 hr. [F-18]3c
2 hr. [F-18]3d
2 hr. [F-18]3e
2 hr. [F-18]3f

Fig. 6

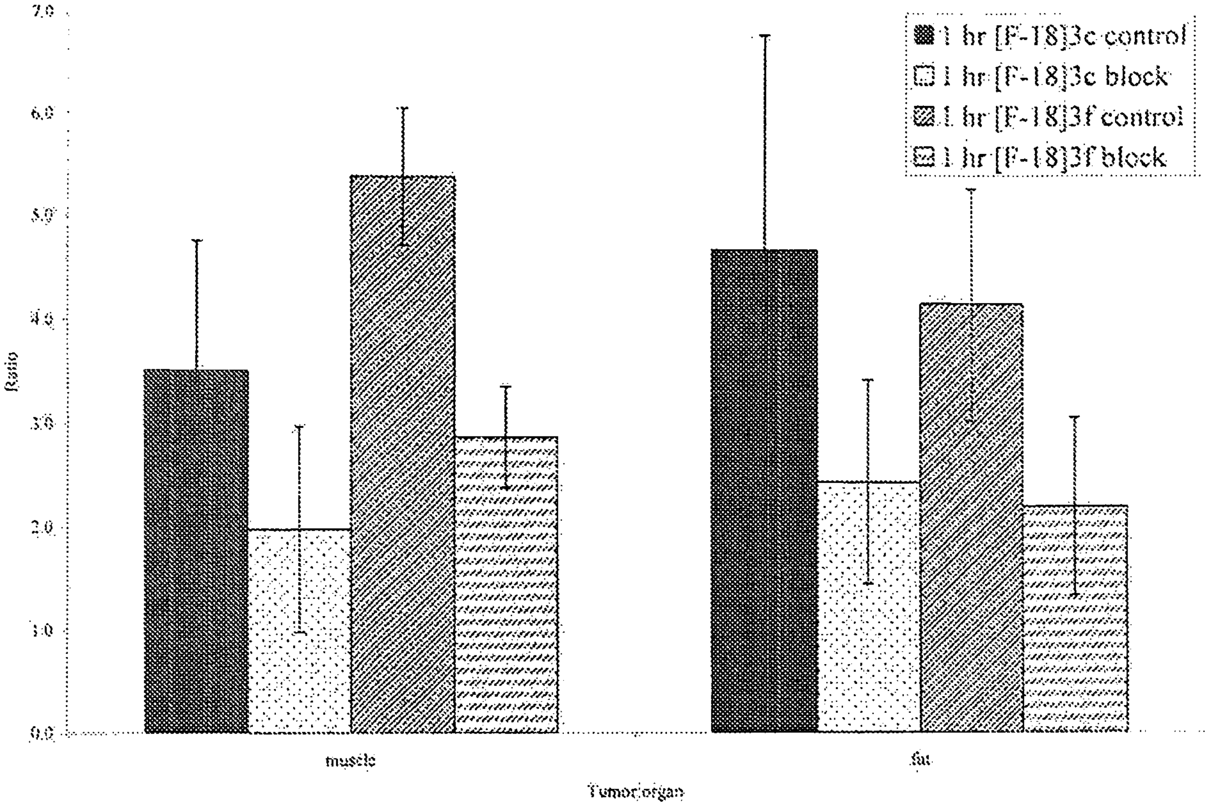

7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
Ratio
1 hr [F-18]3c control
1 hr [F-18]3c block
1 hr [F-18]3f control
1 hr [F-18]3f block
muscle
fat
Tumor/organ

Fig. 7

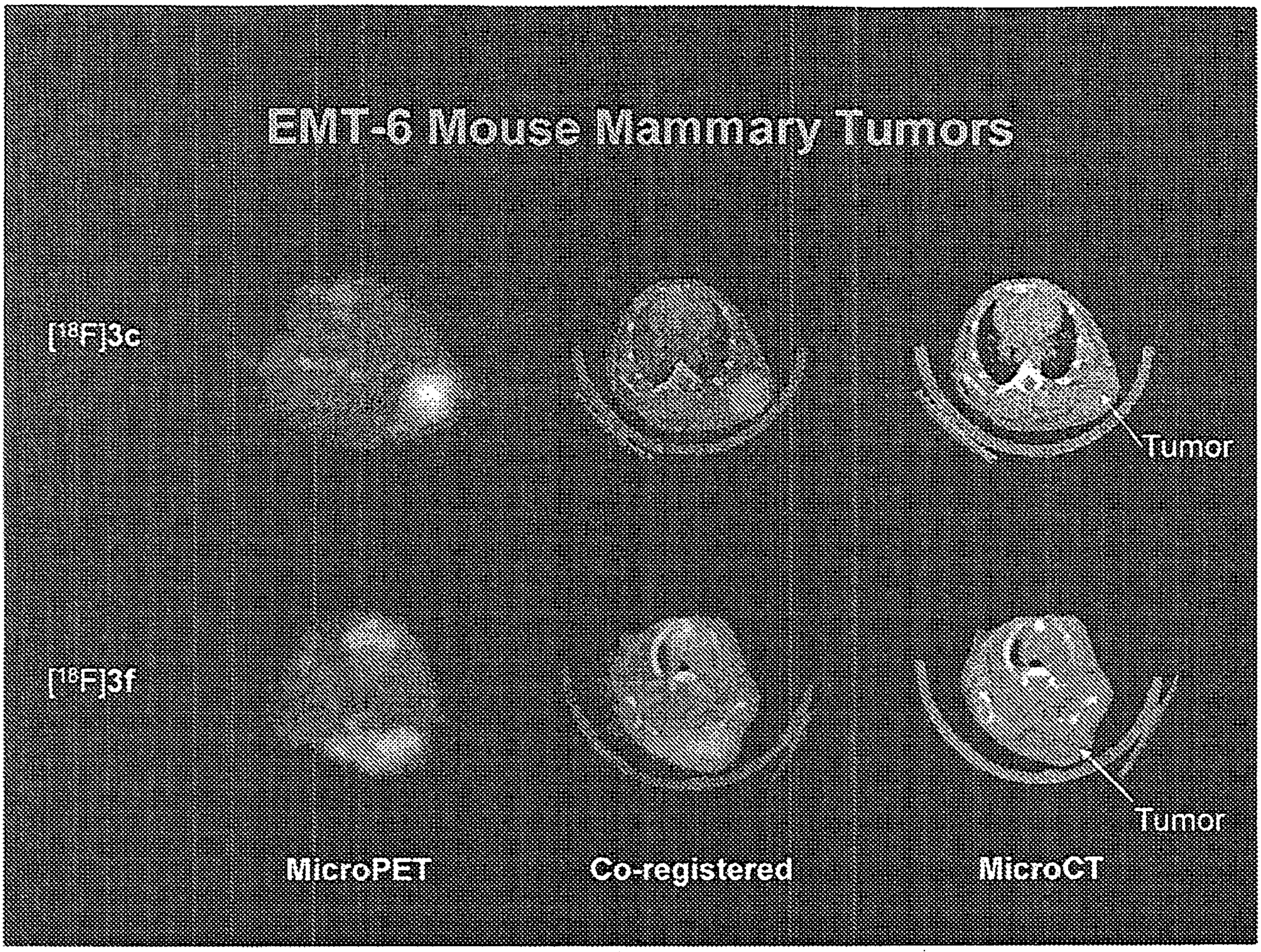

EMT-6 Mouse Mammary Tumors
[18F]3c
Tumor
[18F]3f
Tumor
MicroPET
Co-registered
MicroCT

Fig. 8

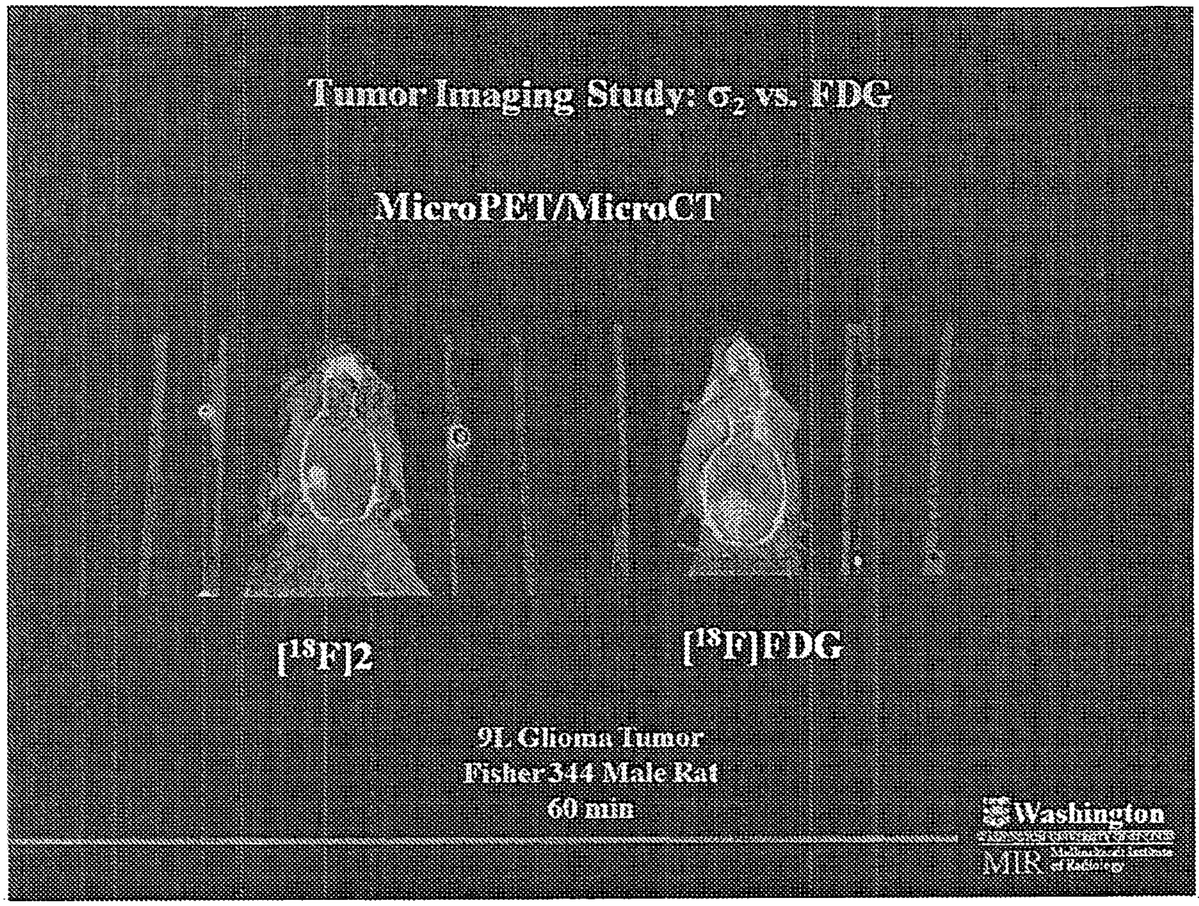

Tumor Imaging Study: σ2 vs. FDG
MicroPET/MicroCT
[18F]2
[18F]FDG
9L Glioma Tumor
Fisher 344 Male Rat
60 min
Washington
MIR

Fig. 9