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(54) Titre : PROCEDE DE RADIOMARQUAGE
(54) Title: RADIOLABELLING PROCESS

(57) Abrégé/Abstract:

The present invention relates to a novel composition comprising 1-amino-3-[¹⁸F]-fluorocyclobutanecarboxylic acid ([¹⁸F]-FACBC) wherein said composition has certain superior properties in comparison with known compositions comprising [¹⁸F]-FACBC. Also provided by the invention is a method to obtain said composition.

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RADIOLABELLING PROCESS

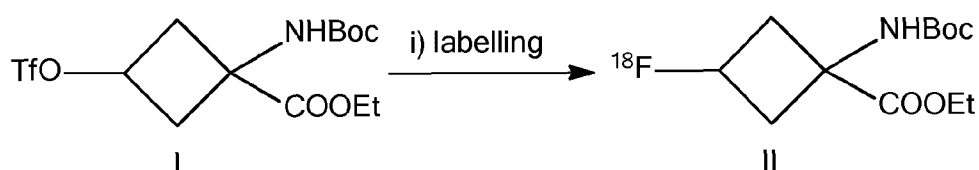
Technical Field of the Invention

The invention relates to a method for the preparation of a radiopharmaceutical compound, in particular an amino acid derivative useful as a positron emission tomography (PET) tracer. The method of the invention is especially suitable when automated and offers advantages over known methods. Particularly, the invention relates to a method for preparation of [^{18}F]-1-amino-3-fluorocyclobutane-1-carboxylic acid ([^{18}F]-FACBC, also known as [^{18}F]-fluciclovine).

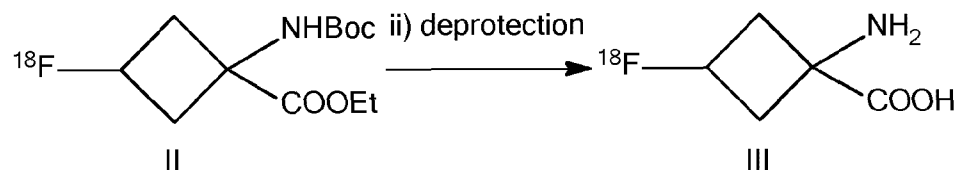
Description of Related Art

The non-natural amino acid [^{18}F]-1-amino-3-fluorocyclobutane-1-carboxylic acid ([^{18}F]-FACBC, also known as [^{18}F]-Fluciclovine) is taken up specifically by amino acid transporters and has shown promise for tumour imaging with positron emission tomography (PET).

A known synthesis of [^{18}F]-FACBC (EP2017258) begins with the provision of the protected precursor compound 1-(N-(*t*-butoxycarbonyl)amino)-3-[[[(trifluoromethyl)sulfonyl]oxy]-cyclobutane-1-carboxylic acid ethyl ester. This precursor compound is first labelled with [^{18}F]-fluoride:



before removal of the two protecting groups:



29925-208

To then obtain injectable [^{18}F]FACBC drug product the crude [^{18}F]FACBC is purified and then formulated.

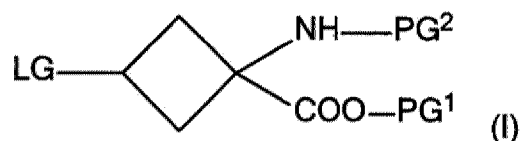
- In the current routine process for producing [^{18}F]FACBC the radiolabelling step (i) is carried out in a reaction vessel followed by transfer of the radiolabelled compound of Formula II above to a tC18 solid phase extraction column for removal of the ester protecting group by alkaline hydrolysis. During this time, the reaction vessel is washed several times with water. The ester-deprotected compound is then returned to the reaction vessel for the removal of the Boc protecting group by acid hydrolysis. Despite washing the reaction vessel several times, the present inventors have determined residual acetonitrile levels in formulated [^{18}F]FACBC drug product ranging from around 100 $\mu\text{g/ml}$ to around 600 $\mu\text{g/ml}$. While these levels are acceptable in terms of permitted daily exposure and in the context of the acceptance criteria for [^{18}F]FACBC drug product, the amount and observed variability is less than ideal.
- There is therefore scope for the provision of an [^{18}F]FACBC drug product wherein the levels of acetonitrile are more tightly controlled, and preferably within a lower concentration range.

Summary of the Invention

- The present invention relates to a novel composition comprising 1-amino-3- ^{18}F -fluorocyclobutanecarboxylic acid ([^{18}F]-FACBC) wherein said composition has certain superior properties in comparison with known compositions comprising [^{18}F]-FACBC. More particularly, the present invention provides an [^{18}F]FACBC composition that has low and consistent amounts of residual solvent. Also provided by the invention is a method to obtain said composition.

In one aspect, the invention relates to a method to obtain a composition comprising 1-amino-3-[^{18}F]-fluorocyclobutanecarboxylic acid ([^{18}F]-FACBC), wherein said composition comprises acetonitrile (MeCN) at a concentration of no greater than 50 $\mu\text{g/mL}$, wherein said method comprises:

- 5 (i) reacting in a reaction vessel [^{18}F]fluoride with a precursor compound of Formula (I):



wherein:

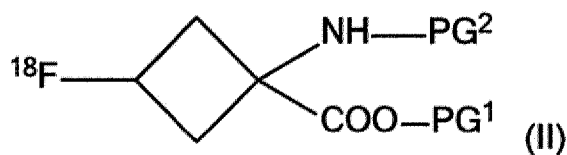
LG is a leaving group;

- 10 PG¹ is carboxy protecting group; and

PG² is an amine protecting group;

wherein said reacting step is carried out in acetonitrile;

to obtain a reaction mixture comprising a compound of Formula (II):

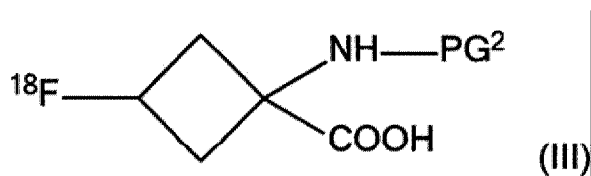


- 15 wherein:

PG¹ and PG² are as defined for Formula (I);

(ii) transferring said compound of Formula (II) out of said reaction vessel onto a cartridge to leave the reaction vessel empty;

(iii) applying heat to the empty reaction vessel whilst at the same time carrying out removal of PG¹ and thereby obtain a compound of Formula (III):



wherein PG² is as defined for Formula (I); and

- 5 (iv) transferring the compound of Formula (III) back into said heat treated empty reaction vessel to carry out removal of PG² and thereby obtain [¹⁸F]-FACBC.

Detailed Description of Preferred Embodiments

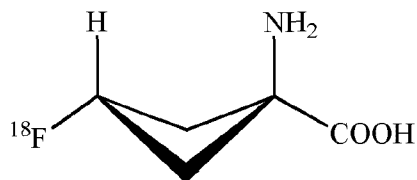
- In one aspect the present invention relates to a composition comprising 1-amino-3-
 10 [¹⁸F]-fluorocyclobutanecarboxylic acid ([¹⁸F]-FACBC) wherein said composition comprises acetonitrile (MeCN) at a concentration of no greater than 50 µg/mL.

In one embodiment the composition of the present invention comprises MeCN at a concentration no greater than 20 µg/mL.

- In one embodiment the composition of the present invention has a radioactive concentration (RAC) of between 500-5000 MBq/ml, preferably between 1000-5000 MBq/ml. The RAC of the composition of the present invention is preferably the RAC of the drug product as soon as this is obtained, i.e. immediately following radiofluorination, deprotection, purification and formulation.

In one embodiment the composition of the present invention has a radiochemical purity (RCP) of at least 99%.

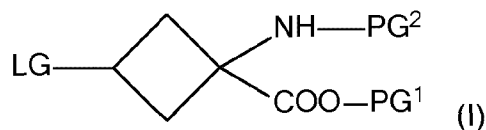
- 10 In one embodiment said [^{18}F]FACBC in the composition of the present invention is trans-1-amino-3- ^{18}F -fluorocyclobutanecarboxylic acid (*anti*- ^{18}F -FACBC):



The composition of the invention is preferably obtainable by the method of the invention described hereinbelow.

- 15 In another aspect, the present invention provides a method to obtain the composition as defined above wherein said method comprises:

(i) reacting [^{18}F]fluoride with a precursor compound of Formula I:



wherein:

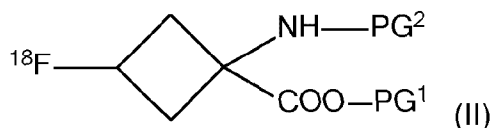
- 20 LG is a leaving group;

PG¹ is carboxy protecting group; and,

PG² is an amine protecting group;

wherein said reacting step is carried out in acetonitrile;

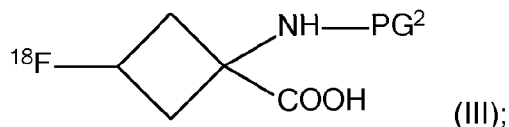
to obtain a reaction mixture comprising a compound of Formula II:



5 wherein:

PG¹ and PG² are as defined for Formula I;

(ii) transferring said compound of Formula II out of said reaction vessel to carry out removal of PG¹ and thereby obtain a compound of Formula III:



10 wherein PG² is as defined for Formula I;

(iii) simultaneously to step(ii) applying heat to said reaction vessel;

(iv) transferring said compound of Formula III back into said reaction vessel to carry out removal of PG² and thereby obtain [¹⁸F]-FACBC.

The method of the invention is largely carried out as described in the art (e.g. Shoup *et al* 1999 J Labelled Comp Radiopharm; 42: 215-225, Svadberg *et al* 2011 J Labelled Comp Radiopharm; 55: 97-102) with the addition of step (iii).

The “[¹⁸F]fluoride” suitable for use in the method of the invention is normally obtained as an aqueous solution from the nuclear reaction ¹⁸O(p,n)¹⁸F. In order to increase the reactivity of fluoride and to reduce or minimise hydroxylated by-products resulting from the presence of water, water is typically removed from [¹⁸F]-fluoride prior to the reaction, and fluorination reactions are carried out using

- anhydrous reaction solvents (Aigbirhio *et al* 1995 J Fluor Chem; 70: 279-87). A further step that is used to improve the reactivity of [¹⁸F]-fluoride for radiofluorination reactions is to add a cationic counterion prior to the removal of water. Suitably, the counterion should possess sufficient solubility within the anhydrous reaction solvent to maintain the solubility of the [¹⁸F]-fluoride. Therefore, counterions that are typically used include large but soft metal ions such as rubidium or caesium, potassium complexed with a cryptand such as KryptofixTM, or tetraalkylammonium salts, wherein potassium complexed with a cryptand such as KryptofixTM, or tetraalkylammonium salts are preferred.
- 5
- 10 A “precursor compound” comprises a non-radioactive derivative of a radiolabelled compound, designed so that chemical reaction with a convenient chemical form of the detectable label occurs site-specifically; can be conducted in the minimum number of steps (ideally a single step); and without the need for significant purification (ideally no further purification), to give the desired radiolabelled
- 15 compound. Such precursor compounds are synthetic and can conveniently be obtained in good chemical purity.

A suitable “leaving group” in the context of the present invention is a chemical group that can be displaced by nucleophilic displacement reaction with fluoride ion. These are well-known in the art of synthetic chemistry. In some

20 embodiments the leaving group of the present invention is a linear or branched C₁₋₁₀ haloalkyl sulfonic acid substituent, a linear or branched C₁₋₁₀ alkyl sulfonic acid substituent, a fluorosulfonic acid substituent, or an aromatic sulfonic acid substituent. In other embodiments of the invention the leaving group is selected from methanesulfonic acid, toluenesulfonic acid, nitrobenzenesulfonic acid,

25 benzenesulfonic acid, trifluoromethanesulfonic acid, fluorosulfonic acid, and perfluoroalkylsulfonic acid. In some embodiments the leaving group is either methanesulfonic acid, trifluoromethanesulfonic acid or toluenesulfonic acid and in another embodiment the leaving group is trifluoromethanesulfonic acid.

The term “protecting group” refers to a group which inhibits or suppresses undesirable chemical reactions, but which is designed to be sufficiently reactive

30

that it may be cleaved from the functional group in question to obtain the desired product under mild enough conditions that do not modify the rest of the molecule.

Protecting groups are well known to those skilled in the art and are described in 'Protective Groups in Organic Synthesis', Theodor W. Greene and Peter G. M.

5 Wuts, (Fourth Edition, John Wiley & Sons, 2007).

The PG¹ "carboxy protecting group" is preferably linear or branched C₁₋₁₀ alkyl chain or an aryl substituent. The term "alkyl" used either alone or as part of another group is defined as any straight, branched or cyclic, saturated or unsaturated C_nH_{2n+1} group. The term "aryl" refers to any C₆₋₁₄ molecular fragment

10 or group which is derived from a monocyclic or polycyclic aromatic hydrocarbon, or a monocyclic or polycyclic heteroaromatic hydrocarbon. In one embodiment of the method of the invention PG¹ is selected from methyl, ethyl, t-butyl and phenyl.

In another embodiment of the invention PG¹ is methyl or ethyl and in yet another embodiment PG¹ is ethyl.

15 The PG² "amine protecting group" suitably prevents reaction between ¹⁸F and the amino group in the process of providing the compound of Formula II. Examples of suitable amine protecting groups include various carbamate substituents, various amide substituents, various imide substituents, and various amine substituents.

Preferably, the amine protecting group is selected from the group consisting of
20 linear or branched C₂₋₇ alkyloxycarbonyl substituents, linear or branched C₃₋₇ alkenyloxycarbonyl substituents, C₇₋₁₂ benzyloxycarbonyl substituents that may have a modifying group, C₂₋₇ alkyldithiooxycarbonyl substituents, linear or branched C₁₋₆ alkylamide substituents, linear or branched C₂₋₆ alkenylamide substituents, C₆₋₁₁ benzamide substituents that may have a modifying group, C₄₋₁₀

25 cyclic imide substituents, C₆₋₁₁ aromatic imine substituents that may have a substituent, linear or branched C₁₋₆ alkylamine substituents, linear or branched C₂₋₆ alkenylamine substituents, and C₆₋₁₁ benzylamine substituents that may have a modifying group. In some embodiments of the invention PG² is selected from t-butoxycarbonyl, allyloxycarbonyl, phthalimide, and N-benzylideneamine. In other

30 embodiments PG² is selected from t-butoxycarbonyl or phthalimide. In one

embodiment of the invention PG² is t-butoxycarbonyl.

The term “reacting” refers to bringing two or more chemical substances (typically referred to in the art as “reactants” or “reagents”) together to result in a chemical change in one or both/all of the chemical substances.

- 5 The “removal of PG¹” is carried out using a reagent capable of removing the carboxy protecting group PG¹ from the compound of Formula II during step (ii) of the method of the invention. Suitable such carboxy deprotecting agents are well-known to the skilled person (see Greene and Wuts, *supra*) and may be either an acid or an alkaline solution. The concentration of the PG¹ deprotecting agent is
- 10 not limited as long as it is sufficient to remove the carboxy protecting group PG¹ and does not have an effect on the final purity or results in an incompatibility with any container used. Preferably the PG¹ deprotecting agent is an alkaline solution. In certain embodiments the PG¹ deprotecting agent is a sodium hydroxide or a potassium hydroxide solution and in a preferred embodiment is a sodium
- 15 hydroxide solution, for example of 0.5-2.0M. The reacting step is enabled by closing the outlet of the SPE column so that the PG¹ deprotecting agent is retained therein for a specified amount of time. The temperature and the duration of this reacting step need to be sufficient to permit removal of the PG¹ carboxy deprotecting group. In certain embodiments the reacting step is carried out at
- 20 room temperature and for a duration of between 1-5 minutes.

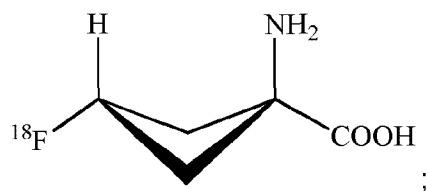
- Step (iii) comprises applying heat to the reaction vessel, which may be carried out using methods well-known to the person skilled in the art and must be suitable for application to the reaction vessel so that the reaction vessel may be used for the subsequent step (iv). This step (iii) is carried out “simultaneously” to step (ii),
- 25 which is to say at the same time as the carrying out removal of PG¹, i.e. after the compound of Formula II has been transferred out of said reaction vessel. A suitable temperature for this heating step should be no greater than the tolerance of the reaction vessel, e.g. for a reaction vessel made from cyclic olefin copolymer (COC) a temperature of no greater than about 130°C and for a reaction vessel
- 30 made from polyetheretherketone (PEEK) a temperature of no greater than about

200°C. For convenience, the temperature used to heat the reaction vessel in step (iii) may be as close as possible to the temperature used during the labelling step (i). For radiolabelling suitable temperatures that are used are in the range of about 80-140°C, in other cases 85-130°C.

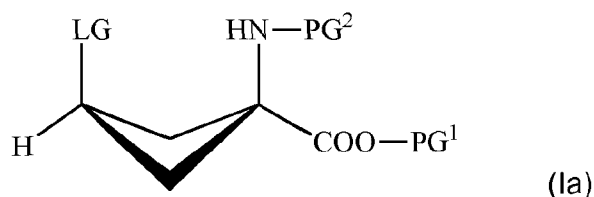
- 5 The "removal of PG²" is carried out with a reagent capable of removing the amine protecting group PG² from the compound of Formula III during the step (iv) of the method of the invention. Suitable such amine deprotecting agents are well-known to the skilled person (see Greene and Wuts, *supra*) and may be either an acid or an alkaline solution. The concentration of the PG² deprotecting agent is not
- 10 limited as long as it is sufficient to remove the carboxy protecting group PG². Preferably the PG² deprotecting agent is an acid solution. A suitable acid preferably includes an acid selected from inorganic acids such as hydrochloric acid, sulfuric acid and nitric acid, and organic acids such as perfluoroalkyl carboxylic acid, e.g. trifluoroacetic acid. In certain embodiments, the PG²
- 15 deprotecting agent is hydrochloric acid, and in other embodiments when HCl is used as PG² deprotecting agent it is at a concentration of 1.0-4.0M. Step (iv) is preferably carried out with heat to allow the removal of PG² reaction to proceed more rapidly. The reaction time depends on the reaction temperature or other conditions. For example, when step (iv) is performed at 60°C, a sufficient reaction
- 20 time is 5 minutes.

Precursor compounds of Formula I may be obtained by following or adapting methods known in the art, such as for example described by McConathy *et al* (2003 Appl Radiat Isotop; 58: 657-666) or by Shoup and Goodman (1999 J Label Comp Radiopharm; 42: 215-225).

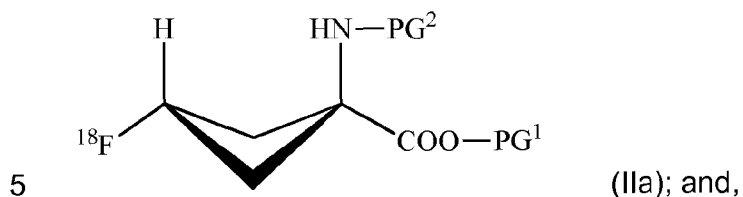
- 25 In a preferred aspect, the [¹⁸F]-FACBC is trans-1-amino-3-[¹⁸F]-fluorocyclobutanecarboxylic acid (*anti*-[¹⁸F]-FACBC):



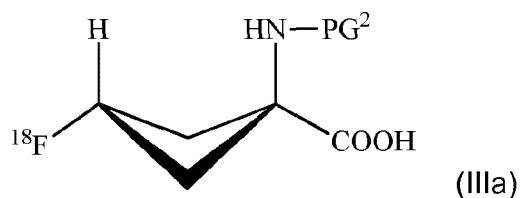
said compound of Formula I is a compound of Formula Ia:



said compound of Formula II is a compound of Formula IIa:



said compound of Formula III is a compound of Formula IIIa:



wherein PG¹ and PG² are as described hereinabove.

In one embodiment, the method of the present invention is automated. Preferably,
 10 the method of the invention is carried out on an automated synthesis apparatus.
 By the term “automated synthesis apparatus” is meant an automated module
 based on the principle of unit operations as described by Satyamurthy *et al* (1999
 Clin Positr Imag; 2(5): 233-253). The term ‘unit operations’ means that complex
 processes are reduced to a series of simple operations or reactions, which can be
 15 applied to a range of materials. Such automated synthesis apparatuses are

preferred for the method of the present invention especially when a radiopharmaceutical composition is desired. They are commercially available from a range of suppliers (Satyamurthy *et al*, above), including: GE Healthcare; CTI Inc; Ion Beam Applications S.A. (Chemin du Cyclotron 3, B-1348 Louvain-La-
5 Neuve, Belgium); Raytest (Germany) and Bioscan (USA).

A commercial automated synthesis apparatus also provides suitable containers for the liquid radioactive waste generated as a result of the radiopharmaceutical preparation. Automated synthesis apparatuses are not typically provided with radiation shielding, since they are designed to be employed in a suitably
10 configured radioactive work cell. The radioactive work cell provides suitable radiation shielding to protect the operator from potential radiation dose, as well as ventilation to remove chemical and/or radioactive vapours. The automated synthesis apparatus preferably carries out the radiosynthesis by means of a cassette. By the term "cassette" is meant a piece of apparatus designed to fit
15 removably and interchangeably onto an automated synthesis apparatus, in such a way that mechanical movement of moving parts of the synthesizer controls the operation of the cassette from outside the cassette, i.e. externally. Suitable cassettes comprise a linear array of valves, each linked to a port where reagents or vials can be attached, by either needle puncture of an inverted septum-sealed
20 vial, or by gas-tight, marrying joints. Each valve has a male-female joint which interfaces with a corresponding moving arm of the automated synthesis apparatus. External rotation of the arm thus controls the opening or closing of the valve when the cassette is attached to the automated synthesis apparatus. Additional moving parts of the automated synthesis apparatus are designed to clip
25 onto syringe plunger tips, and thus raise or depress syringe barrels.

The cassette is versatile, typically having several positions where reagents can be attached, and several suitable for attachment of syringe vials of reagents or chromatography cartridges (e.g. for SPE). The cassette always comprises a reaction vessel. Such reaction vessels are preferably 0.5 to 10 mL, more
30 preferably 0.5 to 5 mL and most preferably 0.5 to 4 mL in volume and are

configured such that 3 or more ports of the cassette are connected thereto, to permit transfer of reagents or solvents from various ports on the cassette. Preferably the cassette has 15 to 40 valves in a linear array, most preferably 20 to 30, with 25 being especially preferred. The valves of the cassette are preferably
5 each identical, and most preferably are 3-way valves. The cassettes are designed to be suitable for radiopharmaceutical manufacture and are therefore manufactured from materials which are of pharmaceutical grade and ideally also are resistant to radiolysis.

Preferred automated synthesis apparatuses for use with the present invention
10 comprise a disposable or single use cassette which comprises all the reagents, reaction vessels and apparatus necessary to carry out the preparation of a given batch of radiofluorinated radiopharmaceutical. The cassette means that the automated synthesis apparatus has the flexibility to be capable of making a variety of different radiopharmaceuticals with minimal risk of cross-contamination,
15 by simply changing the cassette. The cassette approach also has the advantages of: simplified set-up hence reduced risk of operator error; improved GMP (Good Manufacturing Practice) compliance; multi-tracer capability; rapid change between production runs; pre-run automated diagnostic checking of the cassette and reagents; automated barcode cross-check of chemical reagents vs the synthesis
20 to be carried out; reagent traceability; single-use and hence no risk of cross-contamination, tamper and abuse resistance.

The following example serves to further illustrate the invention.

Brief Description of the Examples

Example 1 describes a known method to obtain [^{18}F]FACBC.

25 Example 2 describes the method to obtain [^{18}F]FACBC according to the present invention.

List of Abbreviations used in the Examples

BOC	tert-Butyloxycarbonyl
DP	drug product
HLB	hydrophobic-lipophilic balance
K ₂₂₂	Kryptofix 222
5 MeCN	acetonitrile
QMA	quaternary methyl ammonium
RAC	radioactive concentration

Examples

Comparative Example 1: Prior Art Synthesis of [¹⁸F]FACBC

10 1(i) FASTlab Cassette

All radiochemistry was performed on a commercially available GE FASTlab™ with single-use cassettes. Each cassette is built around a one-piece-moulded manifold with 25 three-way stopcocks, all made of polypropylene. Briefly, the cassette includes a 5 ml reactor (cyclic olefin copolymer), one 1 ml syringe and
 15 two 5 ml syringes, spikes for connection with five prefilled vials, one water bag (100 ml) as well as various SPE cartridges and filters. Fluid paths are controlled with nitrogen purging, vacuum and the three syringes. The fully automated system is designed for single-step fluorinations with cyclotron-produced [¹⁸F]fluoride. The FASTlab was programmed by the software
 20 package in a step-by-step time-dependent sequence of events such as moving the syringes, nitrogen purging, vacuum, and temperature regulation. Vial A contained K₂₂₂ (58.8 mg, 156 μmol), K₂CO₃ (8.1 mg, 60.8 μmol) in 79.5% (v/v) MeCN_(aq) (1105 μl). Vial B contained 4M HCl (2.0 ml). Vial C contained MeCN (4.1ml). Vial D contained the precursor (48.4 mg, 123.5 μmol) in its dry form
 25 (stored at -20°C until cassette assembly). Vial E contained 2 M NaOH (4.1 ml).

The 30 ml product collection glass vial was filled with 200 mM trisodium citrate (10 ml).

1(ii) Production of [^{18}F]Fluoride

No-carrier-added [^{18}F]fluoride was produced via the $^{18}\text{O}(\text{p},\text{n})^{18}\text{F}$ nuclear
5 reaction on a GE PETtrace 6 cyclotron (Norwegian Cyclotron Centre, Oslo).
Irradiations were performed using a dual-beam, 30 μA current on two equal Ag
targets with HAVAR foils using 16.5 MeV protons. Each target contained 1.6
ml of $\geq 96\%$ [^{18}O]water (Marshall Isotopes). Subsequent to irradiation and
delivery to a hotcell, each target was washed with [^{16}O]water (Merck, water for
10 GR analysis). Aqueous [^{18}F]fluoride was passed through the QMA and into the
 $^{18}\text{O}\text{-H}_2\text{O}$ recovery vial. The QMA was then flushed with MeCN and sent to
waste.

1(iii) [^{18}F]Fluoride Labelling

The trapped [^{18}F]fluoride was eluted into the reactor using eluent from vial A
15 and then concentrated to dryness by azeotropic distillation with acetonitrile
(vial C). MeCN was mixed with precursor in vial D from which the dissolved
precursor was added to the reactor and heated to 85° .

1(iv) Removal of Ester Protecting Group

The reaction mixture was diluted with water and sent through the tC18
20 cartridge. Reactor was washed with water and sent through the tC18 cartridge.
The labelled intermediate, fixed on the tC18 cartridge was washed with water,
and then incubated with 2M NaOH after which the 2M NaOH was sent to
waste.

1(v) Removal of BOC Protecting Group

25 The labelled intermediate (without the ester group) was then eluted off the
tC18 cartridge into the reactor using water. The BOC group was hydrolysed by
adding 4M HCl and heating the reactor.

1(vi) Purification

The reactor content with the crude [^{18}F]FACBC was sent through the HLB and Alumina cartridges and into the 30 ml product vial. The HLB and Alumina cartridges were washed with water and collected in the product vial.

5 1(vii) Formulation

2M NaOH and water was added to the product vial, giving a purified drug product (DP) with a total volume of 26 ml.

1(viii) Characterisation

10 Radioactive concentration (RAC) and concentration of acetonitrile were measured in the DP.

FASTlab Run#	RAC (MBq/ml)	MeCN in DP ($\mu\text{g/ml}$)
1	1915	506
2	1804	324
3	1950	302
4	1698	89
5	1570	596
6	1815	218

Example 2: Synthesis of [^{18}F]FACBC using Inventive Method

The method as defined in Example 1 was used except that during removal of the ester protecting group, the empty reactor was heated for 5 minutes.

FASTlab Run#	RAC (MBq/ml)	MeCN in DP ($\mu\text{g/ml}$)
1	3247	16
2	4190	16

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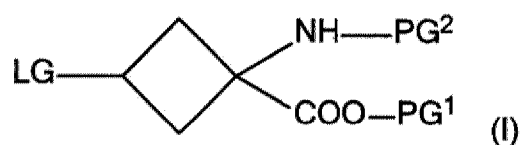
3	1708	16
4	776	17

CLAIMS:

1. A method to obtain a composition comprising 1-amino-3-[^{18}F]-fluorocyclobutanecarboxylic acid ([^{18}F]-FACBC), wherein said composition comprises acetonitrile (MeCN) at a concentration of no greater than 50 $\mu\text{g/mL}$, wherein said

5 method comprises:

(i) reacting in a reaction vessel [^{18}F]fluoride with a precursor compound of Formula (I):



wherein:

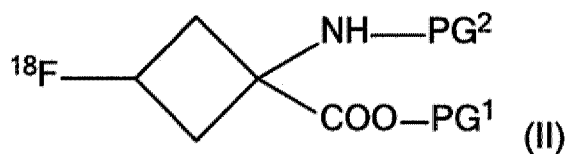
10 LG is a leaving group;

PG¹ is carboxy protecting group; and

PG² is an amine protecting group;

wherein said reacting step is carried out in acetonitrile;

to obtain a reaction mixture comprising a compound of Formula (II):



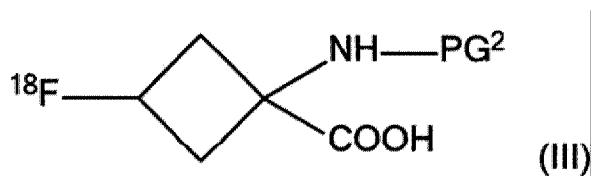
15

wherein:

PG¹ and PG² are as defined for Formula (I);

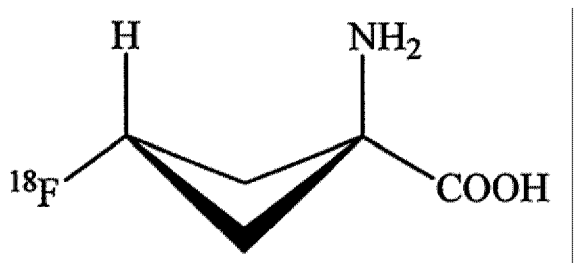
(ii) transferring said compound of Formula (II) out of said reaction vessel onto a cartridge to leave the reaction vessel empty;

(iii) applying heat to the empty reaction vessel whilst at the same time carrying out removal of PG¹ and thereby obtain a compound of Formula (III):

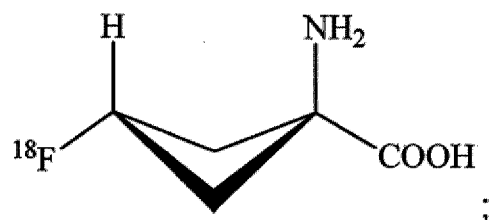


wherein PG² is as defined for Formula (I); and

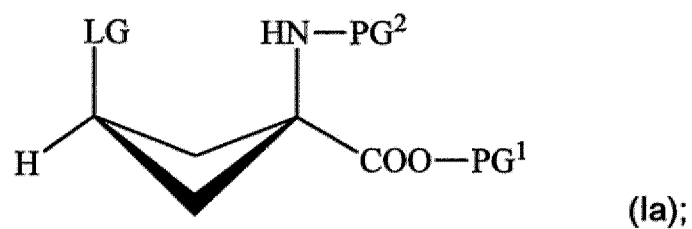
- 5 (iv) transferring the compound of Formula (III) back into said heat treated empty reaction vessel to carry out removal of PG² and thereby obtain [¹⁸F]-FACBC.
2. The method as defined in claim 1, wherein said concentration of MeCN in said composition is no greater than 20 µg/mL.
- 10 3. The method as defined in claim 1 or 2, wherein said composition has a radioactive concentration (RAC) of between 500-5000 MBq/ml.
4. The method as defined in claim 3, wherein said composition has a RAC of between 1000-5000 MBq/ml.
5. The method as defined in any one of claims 1 to 4, wherein said
- 15 composition has a radiochemical purity (RCP) of at least 99%.
6. The method as defined in any one of claims 1 to 5, wherein said [¹⁸F]-FACBC is trans-1-amino-3-[¹⁸F]-fluorocyclobutanecarboxylic acid (*anti*-[¹⁸F]-FACBC):



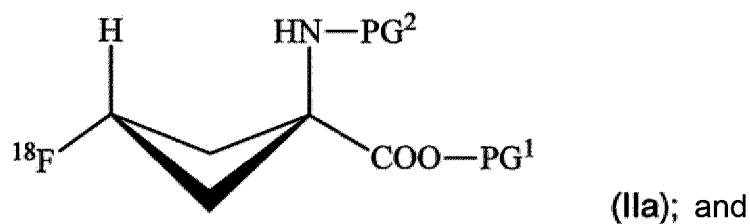
7. The method as defined in any one of claims 1 to 6, wherein LG is a linear or branched C₁₋₁₀ haloalkyl sulfonic acid substituent, a linear or branched C₁₋₁₀ alkyl sulfonic acid substituent, a fluorosulfonic acid substituent, or an aromatic sulfonic acid substituent.
- 5 8. The method as defined in claim 7, wherein LG is methanesulfonic acid, toluenesulfonic acid, nitrobenzenesulfonic acid, benzenesulfonic acid, trifluoromethanesulfonic acid, fluorosulfonic acid, or perfluoroalkylsulfonic acid.
9. The method as defined in claim 7, wherein LG is trifluoromethanesulfonic acid.
- 10 10. The method as defined in any one of claims 1 to 9, wherein PG¹ is a linear or branched C₁₋₁₀ alkyl chain or an aryl substituent.
11. The method as defined in claim 10, wherein PG¹ is methyl, ethyl, t-butyl or phenyl.
12. The method as defined in claim 11, wherein PG¹ is methyl or ethyl.
- 15 13. The method as defined in claim 12, wherein PG¹ is ethyl.
14. The method as defined in any one of claims 1 to 13, wherein PG² is a carbamate substituent, an amide substituent, an imide substituent or an amine substituent.
15. The method as defined in claim 14, wherein PG² is t-butoxycarbonyl, allyloxycarbonyl, phthalimide, or N-benzylideneamine.
- 20 16. The method as defined in claim 15, wherein PG² is t-butoxycarbonyl.
17. The method as defined in any one of claims 1 to 16, wherein said [¹⁸F]-FACBC is trans-1-amino-3-[¹⁸F]-fluorocyclobutanecarboxylic acid (*anti*-[¹⁸F]-FACBC):



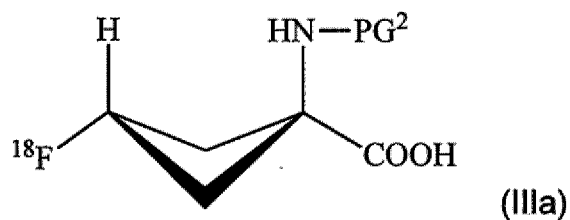
said compound of Formula (I) is a compound of Formula (Ia):



said compound of Formula (II) is a compound of Formula (IIa):



said compound of Formula (III) is a compound of Formula (IIIa):



wherein:

LG is as defined in any one of claims 1 and 7 to 9,

PG¹ is as defined in any one of claims 1 and 11 to 14, and

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PG² is as defined in any one of claims 1 and 14 to 16.

18. The method as defined in any one of claims 1 to 17, which is automated.