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Enantioselective synthesis of no-carrier-added (NCA) 6-[^{18}F]fluoro-L-DOPA

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Abstract

The application of a chiral phase-transfer-catalyst (PTC) in the synthesis of N.C.A. 6-[^{18}F]fluoro-L-DOPA has been recently developed. The 6-trimethylammoniumveratraldehyde triflate precursor and PTC (*O*-allyl-*N*-(9)-anthracenyl-cinchonidinium bromide) were synthesized and successful synthesis route was developed for the preparation of 6-[^{18}F]fluoro-L-DOPA with high radiochemical yields (4–9%, decay uncorrected) and short synthesis time (80 min). The radiochemical purity was over 99% and no D-isomer was detected by HPLC analysis using a chiral mobile phase.

Keywords: 6-[^{18}F]Fluoro-L-DOPA; Phase-transfer; Catalyst preparation; PET

1. Introduction

6-[^{18}F]Fluoro-L-DOPA (6-FDOPA) is the analogue of L-DOPA, which is the biosynthesis precursor for dopamine. As a PET tracer, it has been used for the evaluation of presynaptic dopamine function in cerebral studies in humans (Garnett et al., 1983). Since the early 1980s, many synthetic routes for this radiopharmaceutical have been reported. These approaches are either electrophilic or nucleophilic methods based on the differences of fluoro-labeling agents and labeling reactions (Luxen et al., 1992). In the former approaches, [^{18}F]F_2 or [^{18}F]CH_3COOF have been as the starting radiolabeling agent where fluorine-18 is introduced into the benzene ring via a electrophilic substitution reaction. Although these routes have been widely used for the routine preparation of 6-FDOPA during the past two decades (Luxen et al., 1987, 1990; Firnau et al., 1980; Dolle et al., 1998), several major disadvantages limit use of these electrophilic methods for further application. (a) There include the need of a gas bombard system ([^{20}Ne]neon or [^{18}O]oxygen) to produce [^{18}F]F_2, was needed, which is complicated and expensive. In addition, the [^{18}F]F_2 must be removed from the bombard system by [^{19}F]fluorine gas, so the final products are always carrier-added, and specific activity is low (<2 mCi/mmol). The using of toxic organo-mercuric and tin derivatives as the labeling precursor makes the quality control of the final products difficult (mainly because of the analysis of trace elements. For these obvious reasons, the nucleophilic routes, using [^{18}F]fluoride as the labeling agent via a nucleophilic substitution reaction have become more and more attractive.

Since 6-FDOPA is a chiral compound and precursors containing L-amino-acid structure cannot be labeled with [^{18}F]fluoride directly, the availability of methods to obtain final products with high enantiomeric purity is the key challenge for the nucleophilic synthesis. In the earlier nucleophilic routes, the products at the end of synthesis are racemic, and 6-[^{18}F]fluoro-L-DOPA was separated from its D-isomer by a chiral HPLC column (Lemaire et al., 1990). This additional HPLC purification step, however, largely reduced the yields. To solve this problem, asymmetric synthesis routes have been developed, which can give 6-[^{18}F]fluoro-L-DOPA with

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enantiomeric excess via stereoselective alkylations (Lemaire et al., 1991, 1993; Horti et al., 1995; Najafi, 1995). These multi-step procedures involve the using of sensitive reagents, however, and it is difficult to control the reaction conditions, thus limiting their further application in routine preparation.

Recently, Lemaire et al. (1999) reported a new procedure using a chiral phase-transfer-catalyst (PTC), which avoids the use of sensitive reagents. We describe an efficient alternative procedure which provide 6-[^{18}F]fluoro-L-DOPA in less time.

2. Materials and methods

2.1. Materials

The 4-aminoveratrole, *n*-butyl nitrite, α,α-dichloromethyl methyl ether, cinchonidine, 9-chloromethylanthracene, allyl bromide, K_{222} (Kryptofix222), phenylsilane, iodine, *N*-(diphenylmethylene) glycine *tert*-butyl ester and cesium hydroxide monohydrate were purchased from Acros Organics (Belgium). The methyl trifluoromethanesulfate was purchased from Fluka Chemica Biochemica (Switzerland), and all other chemicals were purchased from Shanghai Chemical Company (Shanghai, China). Chemicals were analytical grade and used without further purification. [^{18}F]Fluoride was produced with a GE PET-trace cyclotron by the ^{18}O(p, n)^{18}F reaction using enriched ^{18}O-Water.

^{1}H-NMR were recorded on an AM-400 NMR spectrometer. Mass Spectra were recorded on an API-2000 LC-MS-MS system (Applied Biosystems Corp.).

2.2. Chromatography system

An LC-10AT HPLC system including a variable wavelength UV detector (Shimadzu, Japan) and a LB 508 Radioflow Detector (EG&G, USA) was used to perform the analytical and purification studies. A $250 \times 4.6\,mm^2$ Shim-lack VP-ODS column (Shimadzu, Japan) was used for the analysis of the labeling products, and was eluted with a mixture of methanol and water (65:35), flow rate 1 ml/min, UV 245 nm or radiodetector. The final products, 6-FDOPA was analyzed with the same column eluted with 0.07 M KH_2PO_4, flow rate 1 ml/min, UV 280 nm or radiodetector; A $300 \times 7.8\,mm^2$ μBondapak C18 semi-preparative column (Waters, Massachusetts, USA) was used for the purification of the final products, by elution with 5 mmol/l sodium acetate, 1 mM EDTA, 17 mM acetic acid and 0.57 mM ascorbic acid, flow rate 4.5 ml/min, UV 280 nm or radiodetector.

Several silica gel (200–300 mesh, Qingdao Haiyang Chemical Factory, China) columns of different sizes were used for the purification of intermediates. A QMA (light) Sep-Pak™ column (Waters, Massachusetts, USA) was used to isolate [^{18}F]fluoride from the enriched [^{18}O]water, eluted with 1 ml of a mixture(77:23) of K_{222} in acetonitrile (10 mg/ml) and K_2CO_3 in water (3 mg/ml). A C18 (plus) Sep-Pak™ column (Waters, Massachusetts, USA) was used for the purification of the labeling products, eluted with CH_2Cl_2.

Thin layer chromatography (TLC) was carried out with glass or terylene plates precoated with GF_{254} silica gel (Sijia Biochemical Plastics Factory, Zhejiang, China). Non-radioactive spots were detected by an UV lamp (Anting Electronic Instrument Factory, Shanghai, China) at 254 nm, and radioactive spots by a γ-ray counter (Rihuan Instrument Factory, Shanghai, China).

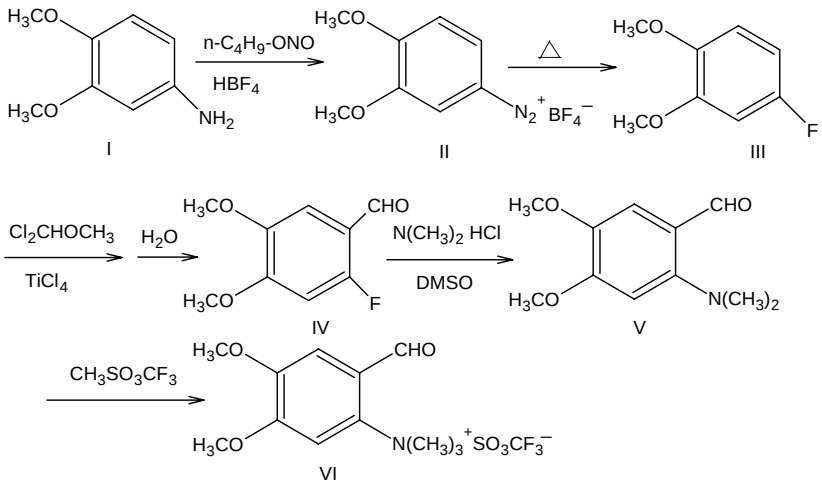
3. Experiments

3.1. Preparation of labeling precursor (VI, 6-trimethylammoniumveratraldehyde triflate)

The labeling precursor was prepared according to Furlanno et al. (1986) and Lemaire et al. (1994). Some of the reaction condition and purification steps were modified and give a higher total yield (~10%) (Scheme 1).

Veratrole-4-diazonium (II). A solution of 10.0 g 4-aminoveratrole (I) in 40.0 ml of fluoroboric acid (aq, 41%) and 40.0 ml of methanol in a 300 ml beaker was cooled to −5°C by an ice–salt bath. The dark purple solution was magnetically stirred, while 10.0 ml of *n*-butyl nitrite was carefully added. During this procedure the temperature of the solution was maintained under 0°C. After stirring for 90 min at −2°C, the solution was diluted with 200 ml of cold ether and was then stirred for another 20 min. The solution was filtered and gave purple colored crystals after standing at −2°C for 2 h. The crystal was washed with 50 ml of cold ether and then dried under vacuum for over 12 h, and veratrole-4-diazonium (II) was obtained (15.0 g, 90%).

4-Fluoroveratrole (III). A 250 ml round-bottom flask containing 20.4 g veratrole-4-diazonium (II) was connected with a long-neck receiving flask which was packed in ice–$CaCl_2$ bath and cooled to −20°C. A water aspirator was connected to the receiving flask to keep the system under low pressure during the reaction. The decomposition of the diazonium salt took place immediately after the 250 ml flask was heated with a Bunsen burner with emission of a white gas. The products of decomposition were directly distilled to the receiving flask during the reaction. To the receiving flask, 10 ml ether was then added, and the solution was washed twice with 5 ml 10% sodium hydroxide and once with 5 ml water. After drying with sodium sulfate, removal of solvent and distillation under reduced pressure, a light yellow liquid (4-fluoroveratrole III, 4.3 g, 35%) was obtained: bp 94–96°C, 10 mm Hg,



Scheme 1. The synthesis route of labeling precursor.

(120–123°C 45 mm Hg, Furlanno et al., 1986); $R_f = 0.35$ (acetone/hexane 25:975); ^{1}H-NMR (CD_3COCD_3): 6.93(m, 1H), 6.78 (m, 1H), 6.62 (t, 1H), 3.80 (d, 6H).

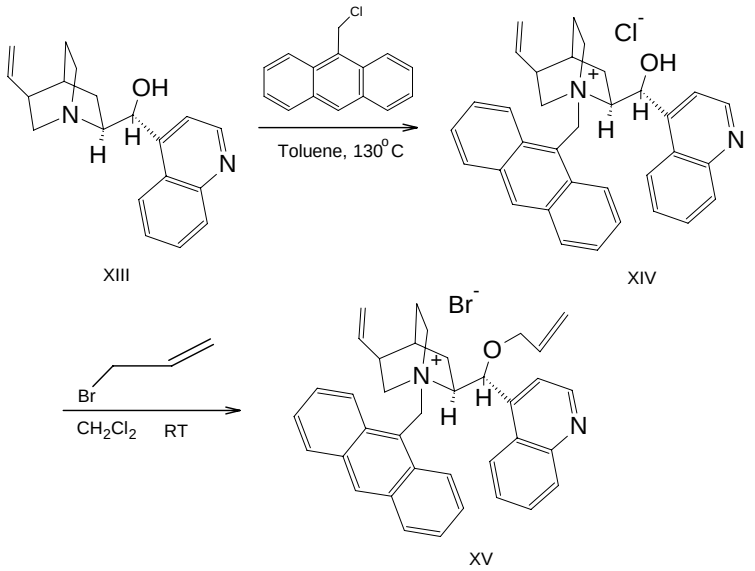
6-Fluoroveratraldehyde (IV). A solution of 5.70 g of 4-fluoroveratrole (III) in 50 ml of methylene chloride was cooled to 0°C with an ice–salt bath under nitrogen atmosphere and then a solution of 7.8 ml titanium tetrachloride in 18 ml of pretreated methylene chloride was dropwise added while stirring. A solution of 6.2 ml of α,α-dichloromethyl methyl ether in 13 ml of methylene chloride was then dropwise added slowly over 20 min. The solution was stirred for 30 min at 0°C and another 4 h at room temperature, while, the color changed from red to dark green. The reaction mixture was poured into a beaker containing 150 g cracked ice. The methylene chloride layer was separated and the water layer was extracted twice with 50 ml of ether. The methylene chloride layer combined with the ether layer was washed twice with 10% sodium bicarbonate and dried with sodium sulfate. After removal of solvent and recrystallization with ether/petroleum ether, a yellow solid (6-fluoroveratraldehyde, IV, 4.01 g, 60%) was obtained: mp 95–97°C (lit. 94–96°C, David C. et al., 1986); $R_f =$ 0.69 (hexane/ether 60:40); ^{1}H-NMR ($CDCl_3$): 10.25 (s, 1H), 7.28 (t, 1H), 6.65 (d, 1H), 3.90 (d, 6H).

6-Dimethylaminoveratraldehyde (V). 6-fluoroveratraldehyde (IV) (3.42 g), dimethylamine hydrochloride (4.60 g) and potassium carbonate (4.53 g) were dissolved with 50 ml of DMSO and 25 ml of water in a 150 ml round-bottom flask. The solution was heated to 180°C with an oil bath while magnetically stirred. After refluxing at this temperature for 3 h, the solution was allowed to cool to room temperature and then was diluted with 100 ml of saturated aqueous potassium carbonate. This mixture was extracted three times with 100 ml of ether, and the organic layer was dried with sodium sulfate. After the solvent was removed, a yellow solid was obtained. The solid was purified by passing through a silica gel column ($2 \times 25\,cm^2$, 200–300 mesh) by elution with hexane/ether 60:40, and gave light yellow solid (6-dimethylaminoveratraldehyde V, 2.77 g, 75%): mp 63–65°C (lit. 62°C, Lemaire et al., 1994); $R_f = 0.40$, hexane/ether 60:40(lit. $R_f = 0.84$, acetonitrile/ether 75:25, Lemaire et al., 1994); ^{1}H-NMR($CDCl_3$): 10.20 (s, 1H), 7.28 (s, 1H), 6.57 (s, 1H), 3.85 (d, 6H), 2.85 (s, 6H).

6-Trimethylammoniumveratraldehyde triflate (VI). A solution of 6-dimethylaminoveratraldehyde (V) (2.77 g) in 25 ml of methylene chloride in three-neck flask was stirred under nitrogen and heated to 40°C with a water bath. To the solution, 7.0 ml of methyl trifluoromethanesulfate was dropwise added. After stirring for 4 h at 40°C, the solution changed from yellow to brown while a precipitate appeared. The solid was collected by filtration and washed with a large volume of cold ether. A white solid (6-trimethylammoniumveratraldehyde triflate, VI, 3.22 g, 64%) was obtained: mp 136–138°C (lit. 140°C, Lemaire et al., 1994); $R_f = 0.40$, acetonitrile/ether 75:25(lit. $R_f = 0.55$ Lemaire et al., 1994); ^{1}H-NMR(D_2O): 9.98 (s, 1H), 7.85 (s, 1H), 7.48 (s, 1H), 4.07 (d, 6H), 3.87 (s, 9H). ESI-MS: calculated for [$C_{13}H_{18}NO_6F_3S$–$SO_3CF_3^-$]: 224.3, found: 224.1.

Preparation of chiral phase-transfer catalyst (O-allyl-N-(9)-anthracenylcinchonidinium bromide). The PTC was prepared according to Corey et al. (1997), and modified as following (Scheme 2).

N-(9)-Anthracenylmethylcinchonidinium chloride (XIV). A 50 ml three-neck flask equipped with a magnetic stirrer, reflux condenser and a nitrogen inlet was charged with a solution of 1.3 g of cinchonidine and 1.0 g of 9-chloromethylanthracene in 15 ml toluene. The mixture was stirred and heated to reflux at 130°C with an oil



Scheme 2. The synthesis route of chiral phase-transfer-catalyst.

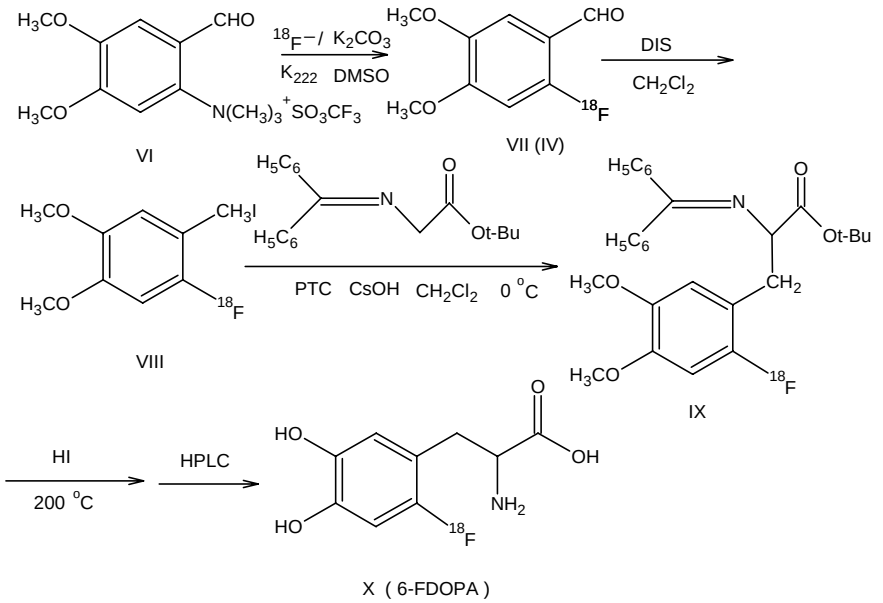
bath for 3 h. The mixture was cooled to room temperature and filtered to give a yellow solid. The solid was washed with toluene and then was recrystallized with chloroform and ether. After dried under vacuum, a light yellow solid (*N*-(9)-Anthracenylmethylcinchonidinium Chloride, XIV, 1.92 g, 79%) was obtained: mp 164–166°C; $R_f = 0.20$ (methanol/ammonia 50:1). ^{1}H-NMR(CDCl3): 9.02(d, 1H), 8.81(t, 2H), 8.68(d, 1H), 8.20(s, 1H), 8.01(d, 1H), 7.95(s, 1H), 7.63(d, 1H), 7.60(d, 1H), 7.54(d, 1H), 7.36(m, 1H), 7.20(m, 4H), 7.05(m, 2H), 6.80(d, 1H), 6.65(d, 1H), 5.41(m, 1H), 5.23(d, 1H), 4.87(d, 1H), 4.70(m, 2H), 4.05(d, 1H), 2.85(t, 1H), 2.39(t, 1H), 2.11(s, 1H), 1.81(m, 2H), 1.68(s, 1H), 1.00–1.20(m, 2H).

O-Allyl-N-(9)-anthracenylcinchonidinium bromide (XV). A three-neck flask equipped with a magnetic stirrer and nitrogen inlet was charged with a solution of 1.8 g of *N*-(9)-Anthracenylmethylcinchonidinium Chloride (XIV) in 40 ml methylene chloride. While being Stirred, 1.6 ml of allyl bromide and 1.2 ml of 50% aqueous sodium hydroxide was added to the solution. The mixture was stirred for another 2 h under room temperature and then was diluted with 25 ml of water. The organic layer was separated and the water layer was extracted twice with 20 ml of methylene chloride. All the organic layers were combined and dried with sodium sulfate and then the solvent was removed to give a yellow solid. After recrystallized with methanol and ether and dried under vacuum, a light orange solid (*O*-allyl-*N*-(9) -anthracenylcinchonidinium Bromide, XV, 0.87 g, 42%) was obtained: mp 169–172°C(lit. 194–196°C, Corey et al., 1997); $R_f = 0.20$ (methanol/ammonia 50:1). ^{1}H-NMR(CD_3OD): 8.88(d, 1H), 8.74(d, 1H), 8.67(d, 1H), 8.42(s, 1H), 8.30(d, 1H), 8.07(m, 3H), 7.78(m, 3H), 7.61(m, 2H), 7.49(m, 2H), 6.80(s, 1H), 6.24(m, 2H), 5.75(d, 1H), 5.53(m, 2H), 5.40(d, 1H), 4.85(m, 2H), 4.36(m, 4H), 3.62(d, 1H), 3.15(m, 1H), 2.75(m, 1H), 2.29(m, 2H), 2.02(m, 1H), 1.82(s, 1H), 1.48(m, 2H); ESI-MS: calculated for $[C_{37}H_{37}N_{2O}Br\text{-}Br]^-$: 525.3, found: 525.5[M-Br $^-$] (Scheme 3).

3.2. Preparation of 6-[^{19}F]fluoro-L-DOPA

6-[^{19}F]Fluoroveratradehyde (VII). A solution of $KF \cdot 2H_2O$ (2.9 mg), K_2CO_3 (1.1 mg) and Kryptofix 222 (20.5 mg) in 100 μl of water was placed in a small reaction vial. The mixture was evaporated to dry under nitrogen flow in an oil bath (140°C). Then 3×100 μl of CH_3CN was added to the residue and evaporated to dryness. A solution of 6-trimethylammoniumveratraldehyde triflate (VI, 20.5 mg) in 1 ml of DMSO was added to the residue. The vial was closed and heated with the oil bath (160°C) for 10 min. The reaction mixture was transferred to a beaker containing 30 ml of 0.5 N HCl and then passed through a C-18 Sep-Pak™ (plus) column and washed in turn with 5 ml of 0.5 N HCl and 10 ml water. At last the column was eluted with 5 ml of CH_2Cl_2, which was dried by passing through a small $MgSO_4$ column. The $MgSO_4$ column was washed with another 5 ml of CH_2Cl_2 and the removal of the solvent gave a yellow solid. After purified with a silica gel column (1×10 cm, 200–300 mesh, fluent: 100% CH_2Cl_2), a light yellow solid (6-fluoroveratradehyde, IV, 2.8 mg, 45%) was obtained. $R_f = 0.53$, 100% CH_2Cl_2 (lit. $R_f = 0.20$); MS: calculated for $[C_9H_9O_3F]$ 184.2 found 184.3 $[M^+]$.

Diiodolane (DIS). DIS was prepared according to the literature (Keinan et al., 1990). 127.8 mg of iodine



Scheme 3. The synthesis route of 6-FDOPA.

was placed in a 5 ml round-bottom flask equipped with a magnetic stirrer. 250 μl of phenylsilane and 15 μl of ethyl acetate was added while stirring at room temperature. The final mixture was used for the iodination reaction immediately without further purification.

2-[^{19}F]Fluoro-4,5-dimethoxybenzyl iodide(VIII). A solution of 30 mg of 6-fluoroveratradehyde in 10 ml of CH_2Cl_2 were added to the previous DIS mixture, and stirred at room temperature for 5 min. Then 0.5 ml of $NaHCO_3$ (10%, aq) and 0.5 ml of $Na_2S_2O_3$ (10%, aq) was added to quench the reaction. After washing with 5 ml of water and drying with anhydrous $MgSO_4$, the organic layer was concentrated and purified on a silica gel column ($1 \times 10\,cm^2$, 200–300 mesh, fluent: 100% CH_2Cl_2) to give a yellow solid (2-fluoro-4,5-Dimethoxybenzyl Iodide, 43.9 mg, 91%): mp 91–93°C; $R_f = 0.88$, 100% Ch_2Cl_2(lit. mp 91°C, $R_f = 0.60$, hexane/ether 25:75, Lemaire et al., 1994); 1H-NMR(CDCl3): 6.80 (d, 1H), 6.58(d, 1H), 4.46(s, 2H), 3.85(m, 6H).

N-(Diphenylmethylene)-2-(2′-[^{19}F]fluoro-4′,5′-dimethoxylbenzyl)-glycine tert-butyl ester(IX). A two-phase mixture of 2-fluoro-4,5-Dimethoxybenzyl Iodide (20.2 mg), the PTC *O*-allyl-*N*-(9)-anthracenylcinchonidinium Bromide (58.8 mg), Schiff base *N*-(diphenylmethylene) glycine *tert*-butyl ester (32.7 mg), cesium hydroxide monohydrate (163.9 mg) and 5 ml of dichloromethane was stirred in a 10 ml round-bottom flask for 10 min at room temperature and then filtrated under vacuum to give a light yellow solution. After removal of the solvent, an impure yellow oil was obtained. This yellow oil was used in the following hydrolysis reaction without further purification. MS: calculated for $[C_{28}H_{30}FNO_4]$: 463.5, found 464.5 [M + H].

6-[^{19}F]Fluoro-l-DOPA (X). The previous yellow oil was mixed with 1.5 ml of hydroiodic acid (freshly distilled on red phosphorous and stabilized with 50 μl of hypophosphorous acid) and 100 mg of red phosphorous in a small vial equipped with a reflux condenser. The vial was heated at 200°C for 20 min in an oil bath. After cooling to room temperature, the mixture was partially neutralized with 1 ml of 6 N NaOH. The resulting mixture was filtrated to remove unsolvable red phosphorous before it was injected into the semi-preparative HPLC column. The retention time was 6.2 min. R_f = 0.70 (CH_3CN:CH_3OH:H_2O 4:1:1); MS, calculated for $[C_9H_{11}FNO_4]$: 215.2, found 214.8.

3.3. Preparation of 6-[^{18}F]fluoro-L-DOPA(X)

[^{18}F]Fluoride production. The no-carrier-added [^{18}F]fluoride was produced by $^{18}O(p, n)^{18}F$ reaction using a small volume of ^{18}O-enriched Water (1.5 ml, >95%) target. The target was bombarded with 16.5 MeV proton beam (25 μA) for 60 min and yielded about 1000 mCi of [^{18}F]fluoride.

6-[^{18}F]Fluoroveratradehyde(VII). 200 mCi of [^{18}F] fluoride in 200 μl of [^{18}O]water was delivered into a small reaction vial containing 22 mg of K_{222} and 3 mg of potassium carbonate. In an oil bath (140°C), the mixture was evaporated to dry under nitrogen flow. Then 3×100 μl of CH_3CN was added to the residue and evaporated to dry. A solution of 6-trimethylammonium-veratraldehyde triflate (VI, 15.2 mg) in 1 ml of DMSO

was added to the residue. The vial was closed and heated with the oil bath (160°C) for 10 min. The reaction mixture was transferred to a beaker containing 30 ml of 0.5 N HCl and then passed through a C-18 Sep-Pak™ (plus) column and washed in turn with 5 ml of 0.5 N HCl and 10 ml water. The column was finally eluted with 5 ml of CH_2Cl_2, which was dried by passing through a small $MgSO_4$ column. The $MgSO_4$ column was washed with another 5 ml of CH_2Cl_2. The 6-[^{18}F]fluoroveratradehyde(VII) was analyzed by HPLC, with a retention time of 3.4 min (condition A) or by TLC, $R_f = 0.70$ (100% CH_2Cl_2).

2-[^{18}F]Fluoro-4,5-dimethoxybenzyl iodide(VIII). To the DIS mixture (as earlier described) the radioactive solution of 6-[^{18}F]fluoroveratradehyde(VII) in CH_2Cl_2 was added, and the mixture was stirred at room temperature for 5 min. Then the mixture was concentrated to about 0.5 ml and purified with a small silica gel column ($1 \times 7.5\,cm^2$, 200–300 mesh) eluted with 5 ml of CH_2Cl_2. A solution of 2-[^{18}F]fluoro-4,5-Dimethoxybenzyl Iodide(VIII) in CH_2Cl_2 was obtained. $R_f = 0.88$ (100% CH_2Cl_2).

N-(Diphenylmethylene)-2-(2′-[^{18}F]fluoro-4′,5′-dimethoxylbenzyl)-glycine tert-butyl ester(IX). The solution was added to a 5 ml round-bottom flask containing 15 mg of *O*-allyl-*N*-(9)-anthracenylcinchonidinium Bromide, 40 mg of cesium hydroxide monohydrate and 2.5 mg of Schiff base *N*-(diphenylmethylene) glycine *tert*-butyl ester, and the mixture was stirred for 5 min at room temperature. After filtrating the filtrate was evaporated to dry under vacuum and the residue was used in the following hydrolysis reaction without further purification.

6-[^{18}F]Fluoro-l-DOPA (X). 1.0 ml of hydroiodic acid (previously distilled on red phosphorous and stabilized with 50 μl of hypophosphorous acid) and 100 mg of red phosphorous was added to the residue in a small vial equipped with a reflux condenser. The vial was heated at 200°C for 20 min with an oil bath. After cooling to room temperature, the mixture was partially neutralized with 0.8 ml of 6 N NaOH. The resulting mixture was filtrated to remove red phosphorous before it was injected into the semi-preparative HPLC column. The retention time was 9.5 min. $R_f = 0.70$ (CH_3CN:CH_3OH:H_2O 4:1:1).

Formulation. Through a sterilizing 0.22-μ membrane filter (Millex-GS, Millipore, Molsheim, France), the HPLC fraction (about 5 ml) was collected into a small vial containing NaCl (50 mg) and ascorbic acid (20 mg).

4. Results and discussion

Labeling precursor. The overall yield of the synthesis of labeling precursor 6-trimethylammonium veratraldehyde triflate was 10–12%, about 100% higher than the reported (Furlanno and Kirk, 1986; Lemaire et al., 1994). This precursor have several advantages compared with the nitro precursors (Lemaire et al., 1994), but unfortunately, it is not stable for long period even stored at 0–4°C. After stored for 6 months, the yields of labeling reaction significantly reduced to lower than 10%.

Chiral phase-transfer catalyst. This chiral PTC was prepared according to the route reported by Corey et al. (1997) with a yield of 32%. To obtain pure products, iterative recrystallization was necessary. Unlike the labeling precursor, this compound was very stable and can be used for over a year.

6-[^{18}F]Fluoro-l-DOPA. The radiochemical yield of this multi-step synthesis averaged from 4–9% (EOB, decay uncorrected) or 7–15% (EOB, decay corrected) with a synthesis time of 80–85 min. Some results are summarized in Table 1.

The ^{18}F labeling reaction yields ranged from 10% to 40%, depending on several factors: the purity of precursor, the quality of the [^{18}F]fluoride solution and the amount of water in DMSO. As the precursor is not stable, after storage for several months, the impurities significantly lower the labeling reaction yields. The quality of the radioactive solution of [^{18}F]fluoride

Table 1
Radiosynthesis data

Steps	Radiochemical yield of each step (%)	Cumulated radiochemical yield (%)	Cumulated synthesis time (min)
Removal of water	—	—	10
Labeling and purification	35–40	35–40	30
Reductive iodination and purification	75–85	26–34	40
Alkylation and purification	75–85	20–29	50
Hydrolysis and HPLC purification	20–30	4–9	85

obtained from the QMA column also affects the yields, but the mechanism is yet not clear. Finally, the water existing in the reaction solvent (DMSO) should be as little as possible. The DMSO used in our lab is 'extra dry' (water <50 ppm). When the precursor of high purity was used, a yield of 30–35% is expected, a little lower than the reported results (Lemaire et al., 1994).

High yield ($>75\%$) of reductive iodination can be easily achieved. We have made an improvement in the purification step by using a small self-made silica gel column (something like a lengthened Sep-Pak column). Using such a column, less time (2 min) and less solvent (5–6 ml) are needed.

The alkylation reaction using chiral phase-transfer catalyst is much more convenient than by those methods previously reported (Lemaire et al., 1993, 1994), and the yield is high ($>75\%$). We also made an improvement here by passing the reaction mixture through a small Sep-Pak™ silica gel column (Waters, USA). This step will obviously reduce the residue after removal of solvent, which may adsorb the products of hydrolysis.

The hydrolysis and HPLC purification steps in our facility are not as successful as expected partly because of the shorter semi-preparative HPLC column (300 mm). Efforts to improve the yields are still in progress.

Radiochemical and chemical purity were assessed by an ODS analytical HPLC column. The radiochemical purity is higher than 99%, and no major unwanted chemicals was observed at two UV wavelengths (220 and 280 nm). Enantiomeric purity was assessed using the same column with a chiral mobile phase. The purity of L-isomer is higher than 95% (90% excess).

After formulation step, the 6[^{18}F]fluoro-L-DOPA solution is ready for animal injection. PET images of a pig showed the expected concentration in striatum.

5. Summary and conclusions

N.C.A. 6-[^{18}F]fluoro-L-DOPA were synthesized via a multi-step procedure, which involves the nucleophilic substitution, DIS reductive iodination, phase-transfer catalytic alkylation and HI hydrolysis reaction. Though the overall radiochemical yield is low (7–15% decay corrected, lit. 25%, Lemaire et al., 1999), the synthesis time was successfully reduced to 80 min (lit. 90 min, Lemaire et al., 1999) by some improvements of purification steps. And the use of microwave heating technique in labeling and hydrolysis reactions is in progress; this may reduce the synthesis time to <60 min. The labeling precursor and chiral phase-transfer catalyst used in the synthesis were also described.

References

Corey, E.J., Xu, F., Noe, M.C., 1997. A rational approach to catalytic enantioselective enolate alkylation using a structurally rigidified and defined chiral quaternary ammonium salt under phase transfer conditions. J. Am. Chem. Soc. 119, 12414–12415.

Dolle, F., Demphel, S., Hinnen, F., et al., 1998. 6-[^{18}F]Fluoro-L-DOPA by radiofluorodestannylation: a short and simple synthesis of a new labeling precursor. J. Labelled Compd. Radiopharm. 41, 105–114.

Firnau, G., Chirakal, R., Sood, S., et al., 1980. Aromatic fluorination with Xeon difluoride: L-3,4-dihydroxy-6-fluorophenylalanine. Can. J. Chem. 58, 1449–1450.

Furlanno, D.C., Kirk, K.L., 1986. An improved synthesis of 4-fluoroveratrole, efficient route to 6-fluoroveratraldehyde and 6-fluoro-D,L-dopa. J. Org. Chem. 51, 4073–4075.

Garnett, E.S., Firnau, G., Nahimias, C., 1983. Dopamine visualized in the basal ganglia of living man. Nature 305, 137–138.

Horti, A., Redmond Jr., D.E., Soufer, R., et al., 1995. No-carrier-added synthesis of 6-[^{18}F]Fluoro-L-DOPA using 3,5,6,7,8,8α-hexahydro-7,7,8α-trimethyl-[6s-(6α,8α,8αβ)]-6,8-methano-2H-1,4-benzoxazin. J. Labeled Compd. Radiopharm. 36, 409–423.

Keinan, E., Perez, D., Sahai, M., et al., 1990. Diiodosilane. 2. A multipurpose reagent for hydrolysis and reductive iodination of ketals, acetals, ketones and aldehydes. J. Org. Chem. 55, 2927–2938.

Lemaire, C., Guillaume, M., Cantineau, R., et al., 1990. No-carrier-added regioselective preparation of 6-[^{18}F]Fluoro-L-Dopa. J. Nucl. Med. 31, 1247–1251.

Lemaire, C., Guillaume, M., Cantineau, R., et al., 1991. An approach to the asymmetric of L-6-[^{18}F]Fluorodopa via nucleophilic fluorination. Appl. Radiat. Isot. 42, 629–635.

Lemaire, C., Plenevaux, A., Cantineau, R., et al., 1993. Nucleophilic enantioselective synthesis of 6-[^{18}F]Fluoro-L-dopa via two chiral auxiliaries. Appl. Radiat. Isot. 44, 737–744.

Lemaire, C., Damhaut, P., Plenevaux, A., et al., 1994. Enantioselective Synthesis of 6-[^{18}F]Fluoro-L-Dopa from no-carrier-added fluoride. J. Nucl. Med. 35, 1996–2002.

Lemaire, C., Guillouet, S., Brihaye, C., et al., 1999. The synthesis of 6-[^{18}F]fluoro-L-DOPA by chiral catalytic phase-transfer alkylation. J. Labelled compd. Radiopharm. 42 (Suppl. 1), s113–s115.

Luxen, A., Bida, G.T., Phelps, M.E., et al., 1987. Synthesis of enantiomarically pure D and L-6-[^{18}F]Fluorodopa and in-vivo metablites via regioselective fluorodemercuration. J. Nucl. Med. 28, 624.

Luxen, A., Perlmutter, M., Bida, G.T., et al., 1990. Remote, semiautomated production of 6-[^{18}F]Fluoro-L-DOPA for human studies with PET. Appl. Radiat. Isot. 41, 275–281.

Luxen, A., Guillaume, M., Melega, W.P., et al., 1992. Production of 6-[^{18}F]Fluoro-L-Dopa and its metabolism in vivo-a critical review. Nucl. Med. Biol. 19, 149–158.

Najafi, A., 1995. Measures and pitfalls for successful preparation of "no carrier added" 6-[^{18}F]Fluoro-L-Dopa from 18F-fluoride ion. Nucl. Med. Biol. 22, 395–397.