High Yield Synthesis of 6-[^{18}F]Fluoro-L-Dopa by Regioselective Fluorination of Protected L-Dopa with [^{18}F]Acetylhypofluorite

Thomas Chaly and Mirko Diksic

Brain Imaging Centre, Montreal Neurological Institute and Hospital; and Department of Neurology and Neurosurgery, McGill University, Montreal, Canada

Regioselective fluorination of a completely protected phosgene derivative of 3,4-dihydrophenyl-L-alanine (5-(benzyl-3',4'-carbonate)-oxazolidine-2,5-dione) with gaseous ^{18}F-labeled acetylhypofluorite and [^{18}F]F_2 in acetonitrile is described. Fluorination with [^{18}F]acetylhypofluorite yields 6-[^{18}F]fluoro-L-dopa with 95% radiochemical purity; fluorination of the same substrate with [^{18}F]F_2 yields a mixture of all three structural isomers in a ratio of 70:16:14 for 6-, 5-, and 2-fluoro compounds. Radiochemical yield, relative to [^{18}F] acetylhypofluorite, measured at the end of the synthesis, is (21 ± 4)% (N = 8). The synthesis requires ~ 40 min (50 min if HPLC was done) and yields the final radiopharmaceutical in a two-step procedure. The specific activity of the final product was ~ 763 mCi/mmol at the end of a 40-min synthesis when 30-min irradiation was used.

J Nucl Med 27:1896–1901, 1986

Fluorine-18- (^{18}F) labeled 6-fluoro-L-dopa (3,4-dihydroxy-6-fluorophenyl-L-alanine) (**4**) has been used as a tracer for assessing the in vivo synthesis of the neurotransmitter dopamine (*1–3*). In several neurological disorders, ^{18}F-labeled metabolites of 6-fluoro-L-dopa accumulated in an area of the brain rich in dopamine (*1–3*). The methods (*4–6*) described to date for the synthesis of ^{18}F-labeled 6-fluoro-L-dopa yield two or three regional isomers of fluorodopa which require time-consuming purification as well as drastic, generally nonquantitative conditions for removal of protecting groups. Each of these operations reduces the final radiochemical yield of the radiopharmaceutical. We recently reported (*7*) a method which gave an increased yield of 6-[^{18}F]fluoro-L-dopa, but involved a difficult synthesis of the starting material. Details are not yet available for a slightly better synthesis recently reported (*8*).

We report a highly regioselective synthesis of 6-[^{18}F]fluoro-L-dopa which does not require high performance liquid chromatography (HPLC) purification since it yields the desired structural isomer almost exclusively. The removal of protecting groups is easy and quantitative even under very mild conditions. The final product contains only small amounts (~5%) of the other two regioisomers when appropriate precautions are taken to exclude moisture. The method, which requires 50 min to complete, produces 6-[^{18}F]fluoro-L-dopa in a radiochemical yield of (21 ± 4)% with a radiochemical purity of (95 ± 2)%.

MATERIALS AND METHODS

All chemicals used were of research purity, obtained from regular suppliers. Broad band proton decoupled ^{19}F-NMR spectra were obtained with a Brucker WP-80 SY spectrometer operated at 75.386 MHz. ^{1}H- and ^{19}F-NMR spectra were obtained with a Varian XL-200 operated at 299.94 MHz and 282.203 MHz, respectively. All chemical shifts are reported relative to an external standard of trichlorofluoromethane ($\delta = 0$) or, in the case of proton spectra, an internal standard of tetramethyl silane.

Fluorination reactions were done using 5% F_2 in nitrogen ("cold" preparations) or 0.5% [^{18}F]F_2 in neon (when labeled compounds were synthesized). Fluorine-18-labeled acetylhypofluorite, the fluorinating agent (*9*), was produced by adapting the method described by

Received Dec. 30, 1985; revision accepted May 9, 1986.

For reprints contact: Mirko Diksic, PhD, Medical Cyclotron Unit, Montreal Neurological Institute, 3801 University St., Montreal, Quebec, H3A 2B4, Canada.

Presented at The Society of Nuclear Medicine Winter Meeting, Metabolic Imaging with NMR, SPECT and PET, March 3–5, 1986, West Palm Beach, FL.

Jewett et al. (*10*). Reagent-grade KOAc . HOAc was used in the reaction with 5% F_2 in nitrogen and $[^{18}F]F_2$ (0.5% F_2 in neon) to produce acetylhypofluorite. The gas mixtures were bubbled through the solution at a rate of 200 ml/min. [The production of $[^{18}F]F_2$ with the mini-medical cyclotron has been described elsewhere (*11*).] All fluorination reactions were done at room temperature. Acetonitrile was dried by refluxing with calcium hydride, and dioxane was dried over KOH for 2–3 days. High performance liquid chromatography was carried out on a reverse phase column* or C-18 Brownlee cartridge†, using radioactivity and uv (λ = 280 nm)† detectors. The elution solvent was 0.1% acetic acid. A radioactivity detector‡ was used with a cell filled with yttrium silicate with a nominal volume of 50 μl.

Thin layer radiochromatography (TLRC) and thin layer chromatography were done on hard layer silica gel§ plates developed in a saturated chamber in an ascended mode. The radiochemical yields are expressed relative to the amount of [^{18}F]acetylhypofluorite measured as total radioactivity present in the reaction vessel at the end of bubbling. (Note that at most, only 50% of ^{18}F ends up in $CH_3COO[^{18}F]F$.)

EXPERIMENT AND RESULTS

Preparation of a Phosgene Derivative of L-Dopa (*2*)

L-Dopa (3,4-dihydroxy-phenyl-L-alanine) [0.985 g (5 mmol)] (**1**) was suspended in 15 ml of dry dioxane or acetonitrile under dry atmosphere. Phosgene was bubbled at a rate of ~5 ml/min through this suspension at room temperature until the solution was clear when reacting in dioxane, or in acetonitrile until most of the L-dopa had reacted (~30 min). After this, dry nitrogen was bubbled through the solution for ~5 min to remove any excess of phosgene. The solution was then filtered through a Millex SR 0.5-μm filter membrane. When reactions were done in dioxane the solvent was removed under reduced pressure using hot air to facilitate the removal of dioxane. A pale yellow viscous liquid was obtained which was found to be very unstable to moisture. It was therefore kept under dry nitrogen until used in the next step. For the fluorination reaction the compound was used without further purification.

In an alternative procedure the reaction with phosgene was done in dry acetonitrile. Any excess of phosgene was removed by bubbling dry nitrogen and the clear solution was filtered through a 0.5-μm teflon membrane in a closed system to avoid contact with moisture. The reaction scheme, outlined in Fig. 1, represents the reaction sequence needed to obtain L-dopa with all functional groups protected (**2**). The ^{1}H-NMR data shows absence of phenolic -OH and carbocylic hydrogen. The integration agrees with the proposed structure of **2**. ^{1}H-NMR data were also compared to those of 4-benzyl-oxazolidine-2,5-dione prepared from phenyl alanine and recrystallized from ethyl acetate-light petroleum (*12*). The infrared (IR) spectra of **2** showed absorption at ν = 1,789 cm^{-1} and 1,746 cm^{-1} corresponding to the carbonyl groups in oxazolidine and carbonate, respectively. The IR also confirmed the absence of free carboxylic acid and phenolic hydroxide absorption. The mass spectrometric data is (m/e, intensity: 249, 4.67 (M^+); 221, 7.19 (M-CO); 205, 10.1 ($M-CO_2$); 149, 37.8 (benzyl-(3,4-carbonate) cation). On the basis of these data and comparison with reactions of other amino acids with phosgene (*13,14*), we concluded that the compound must be 5-(benzyl-3′,4′-carbonate) oxazolidine-2,5-dione (**2**).

Radiofluorination of Protected Dopa with ^{18}F-Labeled Acetylhypofluorite

The pale yellow liquid obtained in the previous step was dissolved in dry acetonitrile. The volume was selected so as to obtain 0.3 to 1 mmol of **2** in 10 ml of the solvent. The solution was transferred into a teflon bubbling tube for fluorination. (When the reaction with phosgene was done in acetonitrile, 10 ml of acetonitrile solution after filtration was used.) Fluorine-18-labeled

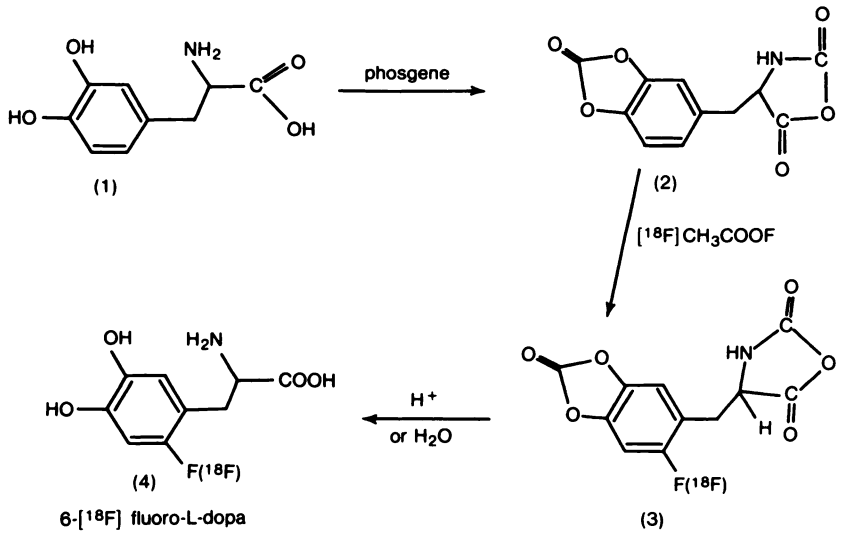

FIGURE 1
Reaction scheme showing synthesis of fluorination substrate (**2**) and 6-[^{18}F]fluoro-L-dopa (**4**)

fluorine gas (on average 0.07 mmol, 0.5% in neon) (*11*) was carried through the KOAc/HOAc column and [^{18}F]acetylhypofluorite produced in the column was bubbled through the solution for 3 min. After this, helium was used at a flow of ~ 300 ml/min to wash the fluorine target and entire fluorine system to transfer as much fluorine as possible into the fluorination vessel.

Fluorination was also done on the mmol scale using CH_3COOF prepared by passing 5% fluorine in nitrogen. The reactions with nonradioactive fluorine were also done with 3–4 times molar excess of (**2**) present during fluorination.

The fluorination product was transferred to a 50 ml round-bottom flask and the solvent removed under reduced pressure using a rotary evaporator. The protecting groups were easily removed by stirring the reaction products with 0.1 N HCl (4 ml) or water (4 ml) for ~10 min. Liquid was removed on a rotary evaporator under reduced pressure using a warm water bath (~80°C). The residue was extracted with a phosphate buffer (pH 7, 2 × 2 ml) and the buffer extract was passed through a SEP-PAK silica cartridge.[¶] An alternative purification was done on a semi-preparative Partisil-M9 column[*] and with 0.1% acetic acid in water as a solvent (*5–7*). This purification removes dopa and 2-[^{18}F]fluoro-dopa from the final radiopharmaceutical. The HPLC-purified radiopharmaceutical showed < 2–3% of 5-fluorodopa when analyzed by the ^{19}F-NMR.

The ^{19}F decoupled NMR spectrum of the product (without HPLC purification) revealed the presence of 6-fluoro-L-dopa (**4**) in ~95% of total products. The 5- and 2-structural isomers of fluorodopa were also observed in the ^{19}F-NMR spectrum. These isomers, however, constituted ~5% of the total product. The final radiopharmaceutical (**4**) was characterized on the basis of its ^{19}F-NMR spectra, and by comparing the retention time on HPLC and the Rf on TLRC with those of an authentic sample prepared by an alternative method (*7*) and with those of another sample supplied by Dr. K. Kirk.[**] After fluorination, TLRC of the crude reaction mixture showed two peaks, one at the origin and the other with Rf = 0.83 using ethyl acetate-hexane (99.5:0.5) as the developing solvent. The ratio between the two spots was ~ 1:1. Because after removal of the protecting groups there was only one spot on TLRC with Rf = 0 in ethyl acetate-hexane (99.5:0.5) as solvent, we believe the spot at the origin was also 6-[^{18}F]fluoro-L-dopa which had lost the protecting groups. The fluorination of **2** had an almost double yield when done in acetonitrile rather than in dioxane.

On HPLC (C-18 cartidge[†]), 6-[^{18}F]fluoro-L-dopa (**4**) had an elution volume of 2.6 ml (k′ = 0.63) using a solution of 0.1% acetic acid in water. The Rf of 6-[^{18}F]fluoro-L-dopa and of an authentic sample of 6-fluoro-L-dopa was 0.71 on hard layer silica gel[§] using methanol-ammonium hydroxide (112:1) as the developing solvent. The chemical shifts and coupling constants agreed well with those previously published (*5–7*).

In a preparation carried out with all ^{18}F radioactivity produced after a 30-min irradiation (*11*), ~55 mCi of [^{18}F]acetylhypofluorite (~56 μmol) yielded 11.6 mCi (~15.2 μmol) of the final radiopharmaceutical. The specific activity of the final product was around 763 mCi/mmol at the end of the synthesis. The specific activity was measured in two independent preparations of 6-[^{18}F]fluoro-L-dopa by HPLC and ~30 independent preparations of CH_3COO[^{18}F]F by titration (*14*). Since we have very good data on the specific activity of CH_3COO[^{18}F]F and there was good agreement with specific activity of the final radiopharmaceutical, we did not always measure specific activity of the final radiopharmaceutical.

Fluorination of **2** with [^{18}F]F_2 is less regioselective, giving a mixture of all three structural isomers in a ratio of 70:16:14 for 6-, 5- and 2-fluoro compounds. Performing the fluorination reaction of **2** in a dry-ice bath did not increase the yield or change the composition of the fluorination mixture. When the amounts of the fluorination substrate (**2**) were varied between 0.3 and 1 mmol, neither of the above-mentioned fluorinations resulted in a significant difference in the yield of the final compound (**2**).

The enantiomeric purity of 6-[^{18}F]fluoro-L-dopa synthesized by this method was compared with that of L-dopa used as a starting material through HPLC, using an in situ generated chiral column (*16*). The synthesis was found to give 6-[^{18}F]fluoro-dopa in an enantiomeric composition similar to that of dopa used as the starting material, a commercial compound of L-dopa. If there is any racemization, it is of the order of 2%–3% as measured by HPLC using the above-mentioned chiral column and solvent described below. The column used in this analysis had k′ = 1.3 and k′ = 4.8, for D- and L-dopa respectively; k′ = 1.4 and k′ = 4.9 for D- and L-fluorodopa, respectively, using 0.05*M* KH_2PO_4, pH = 4.0 buffer containing 1 m*M* $CuSO_4 \cdot 5H_2O$ (*15*). This result confirms an earlier report that hydrolysis of oxazolidine-2,5-dione prepared from optically pure amino acids does not induce appreciable racemization of L-amino acids (*12–14,17,18*) when protecting groups are removed with water, as discussed later.

^{1}H-NMR data for 6-fluoro-L-dopa in D_2O + a few drops of CD_3COOD were as follows:

δ = 6.70 and 6.74 ppm (1H, position-2, AB parts of AB X),

δ = 6.89 and 6.96 ppm (1H, position-5, AB parts of AB X),

δ = 3.80 ppm (1H, X Part of AB X, side chain) with AB parts between 2.94 and 3.1 ppm and 3.14 and 3.2 ppm integrated for one hydrogen each. ^{19}F-NMR in the same solvent:

$\delta = -126.89$ ppm ($^3J_{HF} = 9.9$ Hz, $^4J_{HF} = 7.2$ Hz). A detailed analysis of the NMR spectra agreed well with that described in detail elsewhere (*7*).

DISCUSSION

Fluorination of aromatic compounds using acetylhypofluorite was first reported in 1981 by Rozen et al. (*9*). Fluorination was done in $CFCl_3$ or acetic acid with a reagent generated in situ. Introducing fluorine into aromatic compounds by an electrophilic reaction has recently received special attention because of the interest in ^{18}F-labeled, biologically active compounds, including 6-fluoro-L-dopa (**2**), used for positron emission tomography studies in humans (*1–3*). Several syntheses of 6-[^{18}F]fluoro-L-dopa (**2**) have been reported, but unfortunately all have a low radiochemical yield, require time-consuming HPLC purification, and stringent conditions to remove the protecting groups (*4–6, 8*). The synthesis we recently described (*7*) was 100% regiospecific and produced 6-[^{18}F]fluoro-L-dopa (**2**) in a reasonable yield, but the silane needed as a fluorination substrate is prepared with difficulty, and in scanty amounts. An improvement in the regioselectivity of the fluorination of protected L-dopa giving 2- and 6-structural isomers in equal yields was recently reported, but without details (*8*).

It was reported in an abstract (*8*) that the structural isomer with fluorine in position 5 could be eliminated by using acetate (electron withdrawing group) as protection for the phenolic group in position 4. This gave complete predominance for the directional substitution to the methoxy group in position 3, which directed electrophilic fluorine with an equal probability to positions 2 and 6.

To overcome as many of these problems as possible we have been seeking a protecting group that would deactivate positions 5 and 2 of protected L-dopa. At the same time, we considered that an easy removal of the protecting groups was also important. From the work done in protein synthesis (*12,14,17,18*) and the use of phosgene derivatives of amino acids in gas chromatography (*13*) we realized that phosgene is a reagent that could be used to protect functional groups in L-dopa, and be easily removed after the fluorination reaction is completed.

The reaction of L-dopa (**1**) with phosgene in dioxane at room temperature gave a quantitative yield of the product (**2**), while the reaction in acetonitrile gave a yield of only 60% for the compound (**2**). The intermediary (**2**) was fluorinated with CH_3COO[^{18}F]F and [^{18}F]F_2 in dry dioxane or acetonitrile. Fluorination with [^{18}F]F_2 yields three structural isomers in a ratio similar to that when nonprotected L-dopa is fluorinated with [^{18}F]F_2. Since the reaction of **2** with [^{18}F]F_2 yields three structural isomers, the reaction mechanism is probably different from that involved in fluorination with CH_3COO[^{18}F]F. However, fluorination of partially hydrolyzed compound **2** yielding fluorodopa cannot be ruled out, making the reaction even more complex. Fluorination with [^{18}F]F_2 gave 2-, 5- and 6-[^{18}F]fluoro-L-dopa in a ratio of 14:16:70 (^{19}F-NMR measurement).

Fluorination of **2** with CH_3COO [^{18}F]F in dry acetonitrile or dioxane gave ~95% of 6-[^{18}F]fluoro-L-dopa (**4**) with only small amounts (~5%) of 2- and 5-regioisomers present. The yield of the fluorination products was approximately the same in both solvents investigated. This high regioselectivity could be explained by deactivation of positions 5 and 2. The use of a common electron withdrawing blocking group for the catechol system should also substantially reduce ortho directing influence of both phenolic groups (positions 3 and 4). It was observed earlier that bromination of symmetrically protected dopa and other catechol systems yields only a 6-bromo compound (*7,19*). However, the fluorination of the same symmetrically protected dopa gives all three structural isomers (Diksic M, Chaly T: unpublished data), excluding the possibility of a direct comparison between bromination and fluorination. This symmetrically protected catechol system probably plays a role in the regioselectivity observed in the work reported here, but as mentioned above for 3,4 methoxy dopa, it cannot completely explain the high regioselectivity observed in the fluorination of **2** with acetylhypofluorite.

Since compound **2** could lose some protecting groups very easily (*12,14,17,18*) the 2- and 5-structural isomers are most likely produced from unprotected or partially unprotected L-dopa present in the reaction mixture. The latter could be produced from the protected compound **2** by hydrolysis with traces of water present even in dried solvents. This hypothesis was checked by using a solvent that had not been specially dried; the amounts of 2- and 5-structural isomers were then increased to about 15%, suggesting that water affected the preservation of protected groups in the fluorination substrate.

The specific activity (SA) of the final radiopharmaceutical of ~673 mCi/mmol at the time of injection (being typically ~20 min after the end of synthesis) should be sufficiently high to insure tracer kinetics for 6-[^{18}F]fluoro-L-dopa. (The SA at the end of the synthesis was 770 mCi/mmol.) In a typical study, ~5 mCi of 6-[^{18}F]fluoro-L-dopa would be injected. This will add ~7.4 μ mol[††] of 6-fluoro-L-dopa to the blood which, assuming instantaneous dilution in the plasma, will yield a plasma concentration of about ~ 3.0 μM, much smaller than K_m. The K_m value for dopa is between 300 and 1,400 μM (*20,21*) and the K_m for 6-fluorodopa is 1 μM (*21*). From this it is obvious that the tracer concentration is well below the K_m values for both compounds, thus ensuring behavior of injected

6-[^{18}F]fluoro-L-dopa as a true tracer at the blood-brain barrier.

Our data show that under reaction conditions used in our work there is little, if any, racemization. This corroborates the data of others (*12–14,17,18*) discussed below. The preservation of optical purity in the preparation of oxazolidine-2, 5-diones was also investigated earlier (*12–14,17,18*). Oxazolidine-2, 5-dione was generally prepared by two different routes, by reacting N-carbobenzoxy L-amino acid with PCl_5 (*17,18*) or by reacting free amino acids with $COCl_2$ in dioxane (*12–14*), and subsequently hydrolized back to obtain the original amino acids. The resultant amino acids were shown to have the same optical purity as the starting amino acid (*12–14,17,18*), proving that the synthesis of these substituted oxazolidine-2, 5-diones and subsequent hydrolysis under mild conditions does not induce racemization.

The synthesis of 6-[^{18}F]fluoro-L-dopa reported here produces a greater radiochemical yield than those described previously, takes less time to complete, and gives only the structural isomer desired in a radiochemical yield of (21 ± 4)% (N = 8) with radiochemical purity of (95 ± 2)%. Since the method produces only one structural isomer, elaborate purification is unnecessary. This level of purity is acceptable for radiopharmaceuticals (*22*) labeled with short-lived radionuclides. It should be noted, however, that a very dry condition is essential to obtain this radiochemical purity. For everyday synthesis this requirement may be relaxed and the fluorination mixture passed through a semipreparative HPCL column to insure a proper level of radiochemical purity.

Our results indicate that the reaction of acetylhypofluorite in these solvents involves an electrophilic substitution, which in this particular case, because of the symmetrically protected catechol system and use of common electron withdrawing group for protection of the catechol system, yields the structural isomer desired. This method makes 6-[^{18}F]fluoro-L-dopa (**4**) easily accessible and because of its simplicity facilitates a remotely operated synthesis requiring minimum handling of radioactivity.

FOOTNOTES

* Whatman, Clifton, NJ [Partisil-M9 ODS-2 (10 um, 1.27 × 25 cm)].

† Brownlee Labs Inc., Santa Clara, CA (Brownlee C-18 cartridge).

‡ Isoflo, Nuclear Enterprises, Edinburgh, Scotland.

§ Alltech Associates, Avondale, PA (Uniplates, AN-47521).

†† This point was misquoted in our previous publication (see Ref. *7*).

¶ Waters Associates Inc., Milford, MA (SEP-PAK, Cartridge).

‡‡ An authentic sample was kindly provided to us by Dr. K. L. Kirk, NIH, Bethesda, MD.

ACKNOWLEDGMENTS

This work was supported by a Medical Research Council of Canada (SP-5) grant and the Killam Scholarship Fund of the Montreal Neurological Institute. We appreciate the interest of Dr. W. Feindel and Professor T. H. Chan (Chemistry Department), the editorial help of Dr. V. Lees, and the secretarial help of Ms. C. Elliot.

REFERENCES

1. Garnett ES, Firnau G, Nahmias C: Dopamine visualized in the basal ganglia of living man. *Nature* 305:137–138, 1983
2. Garnett ES, Firnau G, Nahmias C, et al: Striatal dopamine metabolism in living monkeys examined by positron emission tomography. *Brain Res* 280:169–171, 1983
3. Calne DB, Langstron JW, Martin WR, et al: Positron emission tomography after MPTP: Observations relating to the cause of Parkinson's disease. *Nature* 317:246–248, 1985
4. Firnau G, Chirakal R, Sood S, et al: Aromatic fluorination with xenon difluoride. *Can J Chem* 58:1449–1450, 1980
5. Chirakal R, Firnau G, Couse J, et al: Radiofluorination with ^{18}F-labelled acetylhypofluorite: [^{18}F]L-6-fluorodopa. *Int J Appl Radiat Isot* 35:651–653, 1984
6. Firnau G, Chirakal R, Garnett ES: Aromatic radiofluorination with ^{18}F fluorine gas. *J Nucl Med* 25:1228–1233, 1984
7. Diksic M, Farrokhzad S: New synthesis of fluorine-18 labelled 6-fluoro-L-dopa by cleaving the carbon-silica bond with fluorine. *J Nucl Med* 26:1314–1318, 1985
8. Adams MJ, Abeysekera B, Ruth TJ, et al: Synthesis of 6-[^{18}F]fluorodopa using ^{18}F labelled acetylhypofluorite. *J Nucl Med* 26:P125, 1985 (abstr)
9. Rozen S, Lerman O: Acetylhypofluorite as a taming carrier of elemental fluorine for novel electrophilic fluorination of activated aromatic rings. *J Org Chem* 46:4629–4631, 1981
10. Jewett DM, Potocki JF, Ehrenkaufer RE: A preparative gas-solid-phase synthesis of acetylhypofluorite. *Synth Commun* 14:45, 1984
11. Diksic M, Toda Y: Production of ^{18}F-labelled molecular fluorine with a medical mini-cyclotron. *Can J Chem* 61:661–664, 1983
13. König WA, Steinbach E, Ernst K: Phosgene as a reagent for the enantiomeric resolution of 1,2- and 1,3-diols, α-amino alcohols, α-hydroxy acids, and N-methyl- α -amino acids by gas chromatography. *Ang Chem, Int Ed Engl* 23:527–528, 1984
12. Farthing AC: Synthetic polypeptides. Part I. Synthesis of oxazolid-2:5-diones and new reaction of glycine. *J Chem Soc* 3213–3217, 1950
14. Patchornik A, Sela M, Katchalski E: Polytryptophan. *J Am Chem Soc* 76:299–300, 1954 and references therein
15. Diksic M, Jolly D: New high-yield synthesis of ^{18}F-labelled 2-deoxy-2-fluoro-D-glucose. *Int J Appl Radiat Isot* 34:893–896, 1983
16. Grierson JR, Adam MJ: In-situ preparation of a chemically bonded chiral stationary phase for the separation

of aromatic α-amino acid enantiomers. *J Chromatog* 325:103–109, 1985
17. Berger A, Katchalski E: Poly-L-aspartic acid. *J Am Chem Soc* 73:4084–4088, 1951
18. Katchalski E, Sela M: The synthesis and spectrophotometric study of poly-L-tyrosine and poly-3,5-diiodotyrosine. *J Am Chem Soc* 75:5284–5289, 1953
19. Anhoury ML, Crooy P, Deneys R, et al: Bromiated phenylalamines. *Bull Soc Chim Belg* 83:117–132, 1974
20. Firnau G, Sood S, Pantel R, et al: Phenol ionization in dopa determines the site of methylation of catetchol-O-methyltransferase. *Mol Pharmacol* 19:130–133, 1981
21. Kirk KL, Greveling CR: The chemistry and biology of ring-fluorinated biogenic amines. *Med Res Rev* 4:189–220, 1984
22. Krohn KA, Jansholt A-L: Radiochemical quality control of short-lived radiopharmaceuticals. *Int J Appl Radiat Isot* 28:213–227, 1977