High Yield Synthesis of 6-[\(^{18}\)F]Fluoro-L-Dopa

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The radiofluorination of L-dopa with \([^{18}\)F]F\(_2\) was investigated with the purpose of improving the yield of 6-[\(^{18}\)F]fluoro-L-dopa. When boron trifluoride was added to the reaction mixture in hydrogen fluoride (HF), the yield was increased threefold. Nine millicuries of 6-[\(^{18}\)F]fluoro-L-dopa were produced from 100 mCi \([^{18}\)F]F\(_2\) routinely and reliably after 2 hr of preparation. If acetonitrile or water were substituted for HF, little or no 6-fluoro-L-dopa was made.


The cerebral distribution of the neurotransmitter dopamine in humans can be visualized with positron emission tomography and 6-[\(^{18}\)F]fluoro-L-dopa as tracer (1). Using this technique, we have demonstrated the biochemical lesion in patients with unilateral Parkinson’s disease (2,3). Soon it will be possible to measure the kinetics of regional cerebral dopamine in disorders of locomotion and mood (4).

Several syntheses have been described for the tracer 6-[\(^{18}\)F]fluoro-L-dopa (5–7). Thus far, the direct fluorination of L-dopa with [\(^{18}\)F]F\(_2\) in liquid hydrogen fluoride (HF) has proved to be the most reproducible (7). However, moderate radiochemical yield and the need for special equipment to handle liquid HF may be an obstacle to the widespread application of this method.

The chemical study we report here is aimed at improving the radiochemical yield of 6-[\(^{18}\)F]fluoro-L-dopa through a better understanding of the fluorination reaction. We approached this goal in two ways. First, we investigated the effects of the addition of the fluoride acceptors, boron trifluoride (BF\(_3\)), arsenic pentafluoride (AsF\(_5\)), and silicon tetrafluoride (SiF\(_4\)), on the formation of 6-[\(^{18}\)F]fluoro-L-dopa. Second, we tried O-methylated derivatives of L-dopa as substrates. In addition, we studied the direct fluorination reaction in acetonitrile (CH\(_3\)CN) and in water in an attempt to avoid the use of liquid HF as solvent.

MATERIALS AND METHODS

Fluorine-18-(\(^{18}\)F) labeled fluorine gas was produced by the nuclear reaction \(^{20}\)Ne (d,\(\alpha\)) \(^{18}\)F using the 15 meV deuteron beam of the McMaster Tandem Van de Graaff Accelerator (8). The radiofluorination reaction was carried out in an apparatus similar to that described previously (7). The PTFE reaction vessel contained a solution of L-dopa or one of its O-methylated derivatives dissolved in 5.0 ml of either HF, CH\(_3\)CN, or H\(_2\)O, the latter acidified with 1 mmol HCl (Table 1). BF\(_3\), AsF\(_5\), or SiF\(_4\) was then condensed on top of the substrate solution at liquid nitrogen temperature. The amounts of gaseous BF\(_3\), AsF\(_5\), or SiF\(_4\) that were added were determined by measuring the pressure of each gas at a known volume. [\(^{18}\)F]Fluorine gas (190 \(\mu\)mol) diluted with neon (0.5% F\(_2\), 99.5% Ne, Scott Specialty Gases) was then passed for 30 min through each solution of the substrate (470 \(\mu\)mol) either at \(-70^\circ\) for HF, \(-20^\circ\) for CH\(_3\)CN, or 0°C for H\(_2\)O. The amount of \(^{18}\)F retained in the reaction vessel was measured in a calibrated ionization chamber when the bubbling had finished. The amount of \(^{18}\)F retained in the reaction vessel was expressed as a fraction of \(^{18}\)F that had been produced in the target. The amount of \(^{18}\)F in the target was calculated from the energy, intensity of the deuteron beam, time of irradiation, and the known target yield, 91 mCi/\(\mu\)A-h (9).

The solvent—HF, CH\(_3\)CN, or H\(_2\)O—was then evaporated in vacuum and the remainder was dissolved in 10 ml of 0.1M HCl. The HCl was then evaporated and the residue was taken up in 1.5 ml water.

The aqueous reaction mixture was chromatographed with high pressure liquid chromatography (HPLC, stationary phase: two C18 semipreparative columns, 0.78 cm x 30 cm, in series; mobile phase: 0.1% acetic acid in water at 2 ml/min; detector: uv detector at 280 nm and NaI (Ti) scintillation detector to monitor continuously 120 \(\mu\)l of the elute). The
TABLE 1
Effect of Substrate, Solvent, and Lewis Acid on Yield of 6-Fluoro-L-Dopa During Radiofluorination in HF

<table>
<thead>
<tr>
<th>Reaction</th>
<th>Substrate</th>
<th>Solvent</th>
<th>Lewis acid</th>
<th>Fraction of 18F trapped in reaction vessel</th>
<th>Fraction of 18F incorporated in all [18F]fluoro-L-dopas</th>
<th>Isomer distribution (%)</th>
<th>Fraction of 18F in 6-fluoro-dopa</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>L-dopa</td>
<td>HF</td>
<td>—</td>
<td>0.65</td>
<td>0.24</td>
<td>35 5 60</td>
<td>0.14</td>
</tr>
<tr>
<td>2</td>
<td>L-dopa</td>
<td>HF</td>
<td>BF3</td>
<td>0.40</td>
<td>0.34</td>
<td>35 5 60</td>
<td>0.20</td>
</tr>
<tr>
<td>3</td>
<td>L-dopa</td>
<td>HF</td>
<td>AsF5</td>
<td>0.10</td>
<td>0.04</td>
<td>29 16 55</td>
<td>0.02</td>
</tr>
<tr>
<td>4</td>
<td>L-dopa</td>
<td>HF</td>
<td>SiF4</td>
<td>0.33</td>
<td>0.09</td>
<td>38 5 57</td>
<td>0.05</td>
</tr>
<tr>
<td>5</td>
<td>L-dopa</td>
<td>CH3CN</td>
<td>BF3</td>
<td>0.64</td>
<td>0.10</td>
<td>47 39 14</td>
<td>0.01</td>
</tr>
<tr>
<td>6</td>
<td>L-dopa</td>
<td>CH3CN</td>
<td>SiF4</td>
<td>Not determined</td>
<td>Not determined</td>
<td>50 50 0</td>
<td>0</td>
</tr>
<tr>
<td>7</td>
<td>L-dopa</td>
<td>H2O</td>
<td>—</td>
<td>0.50</td>
<td>0.01</td>
<td>56 44 0</td>
<td>0</td>
</tr>
<tr>
<td>8</td>
<td>L-dopa</td>
<td>H2O</td>
<td>BF3</td>
<td>0.60</td>
<td>Not determined</td>
<td>56 44 0</td>
<td>0</td>
</tr>
<tr>
<td>9</td>
<td>3,4-dimethyl-L-dopa</td>
<td>HF</td>
<td>BF3</td>
<td>0.43</td>
<td>0.19*</td>
<td>52 9 39</td>
<td>0.08*</td>
</tr>
<tr>
<td>10</td>
<td>3-O-methyl-L-dopa</td>
<td>HF</td>
<td>—</td>
<td>0.60</td>
<td>0.34</td>
<td>3 54 43</td>
<td>0.14</td>
</tr>
<tr>
<td>11</td>
<td>3-O-methyl-L-dopa</td>
<td>HF</td>
<td>BF3</td>
<td>0.49</td>
<td>0.37</td>
<td>3 54 43</td>
<td>0.16</td>
</tr>
<tr>
<td>12</td>
<td>4-O-methyl-L-dopa</td>
<td>HF</td>
<td>BF3</td>
<td>0.32</td>
<td>0.20</td>
<td>20 26 54</td>
<td>0.11</td>
</tr>
</tbody>
</table>

4.2 mmol of either BF3, SiF4, AsF5 were added.

* Fluorinated 3,4-O-methyl-L-dopa was hydrolyzed by treatment with HBr to obtain [18F]fluoro-L-dopas.

RESULTS

Table 1 summarizes the results of all the experiments performed. It is shown that the highest yield of fluoro-L-dopas was obtained when L-dopa was the substrate, HF was the solvent and BF3 was the Lewis acid. Table 2 shows that the yield of fluoro-L-dopas is enhanced by increasing the amount of BF3 added, up to 4.20 mmol.

Figure 1 illustrates the variety and relative amounts of 18F containing reaction products that were formed under the various reaction conditions. The peak at 16 min comprised 18F containing by-products. The [18F]fluoro-L-dopas eluted at 30 min. More by-products than [18F]fluoro-L-dopas were made when CH3CN with 4.2 mmol BF3 was the solvent (Fig. 1A). The use of HF with 2.1 mmol BF3 reduced the by-products considerably (Fig. 1B). When HF with 4.2 mmol BF3 was used, the number and amount of the by-products were reduced even further. Partial separation between the combined 2- and 5-fluoro-L-dopas and 6-fluoro-L-dopa can be seen (Figs. 1B and C).
TABLE 2
Effect of BF₃ on Yield of Fluoro-L-Dopas During
Radiofluorination of L-Dopa in HF

<table>
<thead>
<tr>
<th>Amount of BF₃ added to reaction mixture (mmol)</th>
<th>Yield of [¹⁸F]fluoro-L-dopas (% of 1 mmol)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>37</td>
</tr>
<tr>
<td>2.08</td>
<td>52</td>
</tr>
<tr>
<td>4.16</td>
<td>84</td>
</tr>
<tr>
<td>4.86</td>
<td>74</td>
</tr>
<tr>
<td>5.98</td>
<td>81</td>
</tr>
</tbody>
</table>

Improved Synthesis Procedure for 6-[¹⁸F]Fluoro-L-Dopa

As the result of the experiment described, we have altered our original method for the preparation of 6-[¹⁸F]fluoro-L-dopa (7) by addition of BF₃ to the reaction mixture. Thus, L-dopa (94 mg, 470 µmol) is dissolved in 5 ml liquid HF in a reaction vessel of the apparatus previously described (7). Boron trifluoride (4.2 mmol) is then condensed into the reaction vessel at -196°C. After this solution has been warmed up to -65°C dilute radioactive fluorine gas (0.5% F₂ in Ne, total amount of F₂ 190 µmol) is passed through it at 80 ml/min. The solvent HF is then evaporated in vacuo at room temperature. From this point, the workup and chromatographic procedure for the isolation of 6-[¹⁸F]fluoro-L-dopa, is identical to that previously described (7).

DISCUSSION

Dilute fluorine has been described as a "highly reactive and correspondingly unselective (fluorinating) reagent" (10). Indeed, when we used it to fluorinate L-dopa we found a variety of fluorination products (Figs. 1A and B). This is not surprising considering the structural complexity of L-dopa and its derivatives. Both are particularly vulnerable to reaction with fluorine because the amino acid side chain can react easily with fluorine radicals. In addition, the substituents of the aromatic ring (—OH, —CH₂R) are electron-donating groups known to promote electrophilic reaction with fluorine. Furthermore, catechols are oxidized readily by F₂. During our investigation, we searched for reaction conditions that have a taming influence on either fluorine or L-dopa. The type of substrate, the reaction solvent, and the Lewis acid all affect isomer distribution and yield.

Effect of Substrate

When O-methylated L-dopas were used as substrates the yield of the 6-fluoro isomer decreased with the extent of the methylation of the hydroxyl groups. Under comparable reaction conditions the fraction of ¹⁸F in 6-fluoro-dopa was highest when unprotected L-dopa was the substrate (Reactions 2, 11, 12, and 9, Table 1). The use of methylated substrates requires the removal of the methyl group (usually by acid hydrolysis for 30 min). This additional time for hydrolysis would lower the practical yield even further. Thus, the protection of the catechol in L-dopa by a methyl group is not a suitable strategy towards a high yield synthesis for 6-fluoro-L-dopa.
Effect of Solvent

When CH3CN-BF3 was used as the solvent instead of HF-BF3, the fraction of 18F in 6-fluoro-L-dopa dropped from 0.20 to 0.01 (Reactions 5 and 2, Table 1). When water was used, no 6-fluoro-L-dopa was formed. The acidity of HF-BF3 (11) is apparently optimal for both a high incorporation of 18F and for the formation of the desired 6-fluoro isomer.

Effect of Fluoride Acceptors

When the Lewis acid, BF3, or AsF5 is added to HF the acidity of the solvent is increased; AsF5 has the greater effect (11). Concomitantly, the two catechol oxygens in dopa can be expected to be protonated (12). Protonated oxygens no longer behave as electron-donating but as electron-withdrawing groups. The latter are known to retard electrophilic aromatic substitution (13). Indeed, we have observed a decreased rate of reaction between F2 and L-dopa or its derivatives as reflected in a lower fraction of 18F that is trapped (Reactions 1, 2, 3, 10, and 11, Table 1). This reduced reaction velocity in HF-BF3 was accompanied by an increase in the yield of the 6-fluoro isomer. The addition of ~4.2 mmol BF3 produced the maximum yield of fluoro-dopas (Table 2).

Unlike BF3 and AsF5, SiF4 does not increase the acidity of HF. Consequently, SiF4 did not increase the yield of the fluoro-L-dopas (Reaction 1 and 4, Table 1). The polar solvents CH3CN-BF3, CH3CN-SiF4, H2O-HCl, and H2O-BF3 not only decreased the yield of the fluoro-L-dopas, they also drastically changed the substitution pattern; fluorne preferentially entered the 2- and 5-position at the expense of the 6-fluoro isomer (Reactions 5 to 8, Table 1).

Lewis acids have been used by many to catalyze electrophilic fluorination reactions. In these reactions they polarize fluorinating agents, like XeF2 (14) and CIF3 (15), to facilitate the electrophilic fluorination. In the present work we believe BF3, AsF5, and SiF4 affect the substrate and not the fluorinating agent F2. This notion is supported by two observations. First, if BF3, AsF5, or SiF4 did interact with F2, the specific activity of the product would be drastically lowered because of isotopic exchange between [18F]F2 and the stable fluoride in a common intermediate. Second, the small size of the fluoride molecule makes it unlikely that the F—F bond in F2 is polarized by fluoride acceptors, such as BF3 or AsF5 (16). The mechanism by which F2 reacts with L-dopa remains to be elucidated.

In their pioneering work, Cacace et al. (10) have warned against interpreting fluorination reactions according to a common reaction mechanism because "the attack of elemental fluoride on aromatic substrates could conceivably involve multiple, and possibly overlapping pathways." We speculate that the fluoro-L-dopas may be formed by electrophilic action of F2 on L-
dopa. Other reactions also occur because we observe a variety of 18F containing reaction products.

We conclude that (a) our original synthesis of 6-[18F]-fluoro-L-dopa (7) can be improved by addition of BF3 to the reaction medium, (b) the solvent HF cannot be replaced without loss in yield, and (c) use of O-methylated L-dopa derivatives as substrates does not increase the yield of 6-fluoro-L-dopa.

We now add 4.2 mmol of BF3 to the solvent HF for the routine production of the radio pharmaceutical 6-[18F]fluoro-L-dopa. Typically, we produce 9 mCi of 6-[18F]fluoro-L-dopa with a specific activity of 220 mCi/mmol from 100 mCi [18F]F2. The procedure takes 2 hr.

FOOTNOTES

* Waters Chromatography Div., Millipor, Milford, MA (μBondapak).
† Bruker Instruments Inc., Billerica, MA (WM-250).

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Note

Work with HF is potentially harmful. Skin contact with even small amounts of HF results in painful chemical burns. Before beginning work with HF, the first-aid treatment procedure (17) should be available and known to all laboratory personnel.

REFERENCES