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Research Article

Synthesis of 2-[^{18}F]fluoroadenosine (2-[^{18}F]FAD) as potential radiotracer for studying malignancies by PET

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Summary
2-[^{18}F]fluoroadenosine (2-[^{18}F]FAD), a potential radioligand for assessment of adenylate metabolism, was synthesized by carrier-added and no-carrier-added procedures via nucleophilic radiofluorination of 2-fluoroadenosine and 2-iodoadenosine. The radiochemical yield, specific radioactivity and radiochemical purity of carrier-added and no-carrier-added 2-[^{18}F]FAD were 5%, 22–30 mCi/μmol and 99%, and 0.5%, 1200–1700 mCi/μmol and 99%, respectively. Copyright © 2006 John Wiley & Sons, Ltd.

Key Words: positron emission tomography; ^{18}F; 2-[^{18}F]fluoroadenosine; cancer; adenylate metabolism

Introduction
Fludarabine phosphate (Fludara, 2-fluoroadenosine-5-phosphate) **1** (Figure 1) is a well-established medication for treatment of leukemia and other types of cancer.[1–12]

Compound **1** is a pro-drug that is rapidly converted in the blood to the free nucleoside 2-fluoroadenosine, **2**[13] (Figure 1) which enters cells and accumulates mainly as the 5'-triphosphate, F-ara-ATP. The mechanism of action of F-ara-ATP is complex and mostly targets DNA and includes incorporation into DNA resulting in repression of further DNA polymerization, inhibition of ribonucleotide reductase and inhibition of DNA ligase and DNA primase.[14]

If radiolabeled with [^{18}F]fluorine, compound **2** would appear to be an interesting candidate for assessing adenylate metabolism by positron emission tomography (PET). Abnormal adenylate metabolism is associated with a

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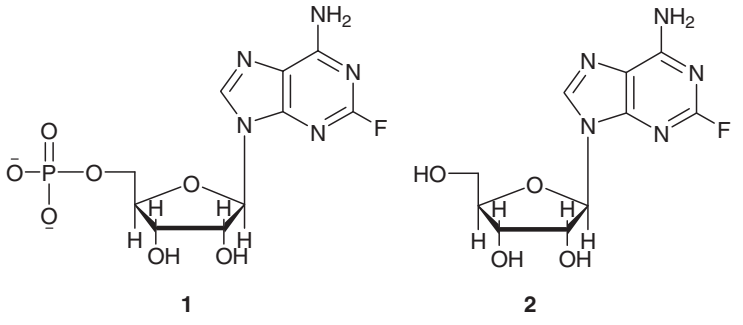

Figure 1. Fludarabine phosphate 1 and 2-fluoroadenosine 2 (F-ara-A)

variety of disorders including cancer, cystic fibrosis, diabetes and cardiac disease.[15] There are currently several radiotracers synthesized having potential for PET imaging of adenylate metabolism (see the latest references in the paper by Mathew *et al.*[16]). Nevertheless, a radiotracer with better imaging properties is needed. Here we present a synthesis of 2-[^{18}F]fluoroadenosine [^{18}F]**2** using a nucleophilic radiofluorination procedure.

Results and discussion

Known methods for the preparation of compound **2** utilize the Schiemann reaction (dediazotisation-fluorination) through the 2,6-diaminopurine riboside, **3** (Figure 2).[17–19] Our attempt to apply the conditions of the Schiemann-type radiofluorination to compound **3** did not reveal detectable amounts of [^{18}F]**2**. In contrast, a substitution of fluorine in the molecule of **2** with [^{18}F]fluoride via the conventional nucleophilic radiofluorination in DMSO solution at 150°C gave radiolabeled [^{18}F]**2**. The optimal heating time was 7 min. The radiofluorination was sluggish at lower temperatures, while at 165–180°C an unidentified by-product prevailed. It is noteworthy that **2** rapidly decomposes under the reaction conditions yielding the by-product. The optimal amount of K_2CO_3 was 0.7 mg, whereas in the presence of larger or smaller amounts of K_2CO_3 lower yields of [^{18}F]**2** were observed (Figure 3). An attempt of carrier-added radiofluoriation in anhydrous acetonitrile gave less than 1% of [^{18}F]**2**. Under optimal radiofluorination conditions the carrier-added [^{18}F]**2** was obtained with radiochemical yield of about 5%, specific radioactivity of 22–30 mCi/μmol and radiochemical and chemical purities greater than 99 and 94%, respectively.

No-carrier-added radiofluorination of 2-iodoadenosine **4** under the same reaction conditions gave the target product [^{18}F]**2** with lower yield (0.1–0.5%), radiochemical and chemical purities greater than 99%, and specific radioactivity of 1200–1700 mCi/μmol. It is likely, that radiofluorination of *O*- and *N*-protected 2-iodoadenosine followed by removal of protective groups should increase the radiochemical yield of [^{18}F]**2**.

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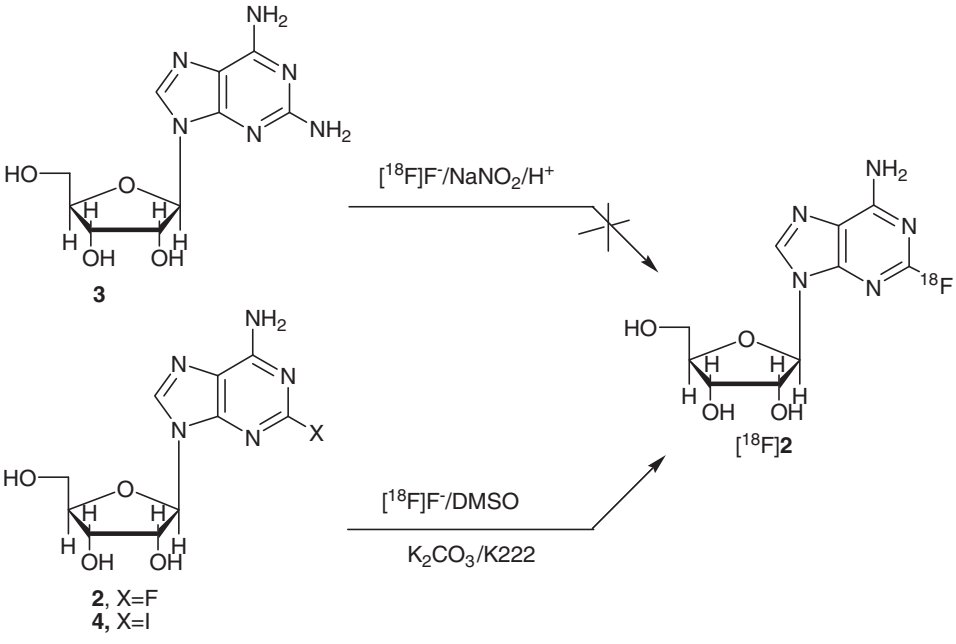

Figure 2. Radiosynthesis of 2-[^{18}F]fluoroadenosine, [^{18}F]2

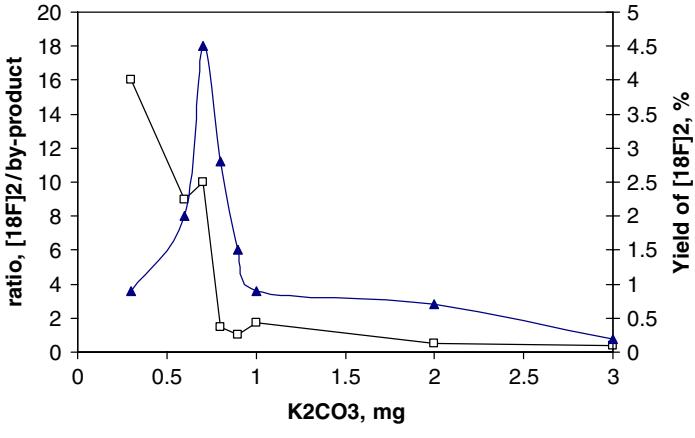

Figure 3. Carrier-added radiosynthesis of [^{18}F]2. Influence of the amount of K_2CO_3 on the radiochemical yield of [^{18}F]2 (filled triangles) and the ratio of [^{18}F]2/[^{18}F]by-product (open squares)

Experimental

Materials and methods

Most reagents and all solvents used were purchased from Aldrich Chemical Co. (Milwaukee, WI). 2-Fluoroadenosine **2** was obtained from SynQuest

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Laboratories, Inc. (Alachua, Florida), and 2-iodoadenosine **4** was obtained from TRC Inc. (North York, Canada). HPLC analysis and purification were performed with two HPLC pumps (model 600/610, Waters, Milford, MA), an in-line Waters UV-detector (254 nm), and a single two-inch NaI crystal flow-count radioactivity detector (Bioscan 3200, Washington, DC). HPLC chromatograms were recorded by a dual channel control/interface module connected to a PC with Galaxy software (Varian). A dose calibrator (model CRC-35R, Capintec, Ramsey, NJ) was used for all radioactivity measurements. [^{18}F]Fluoride was prepared using a GE PETtrace cyclotron. The radiofluorination was performed using an automated radiochemistry module (MicroLab, GE).

Radiochemistry

*2-[^{18}F]fluoroadenosine ([^{18}F]**2**): carrier-added radiosynthesis.* An aqueous solution of [^{18}F]fluoride (prepared by 16 MeV proton irradiation of 98% enriched $H_2^{18}O$), 25 mg of Kryptofix 222, and 0.7 mg potassium carbonate was added to a 10 ml reaction vessel. The mixture was heated at 120–130°C under a stream of argon while water was evaporated azeotropically using addition of acetonitrile. A solution of compound **2** (2 mg) in anhydrous dimethylsulfoxide (0.9 ml) was added to the reaction vessel and heated at 150°C for 7 min. The reaction mixture was cooled, diluted with 1 ml of 0.1% aqueous acetic acid, injected onto a semi-preparative Hamilton PRP-1 HPLC column, 10 μm, 7 × 305 mm (Reno, NV) and eluted with a mixture of CH_3CN:0.1 M ammonium formate 3.5:96.5 at a flow rate of 6 ml/min. The radioactive peak with a retention time of 10 min corresponding to [^{18}F]**2** was collected, the solvent was removed on a rotary evaporator (60–80°C), and the final product was formulated with 5 ml of saline.

An aliquot of the final solution of known volume and radioactivity was applied to a Hamilton PRP-1 HPLC column, 10 μm, 7 × 305 mm. A mobile phase (CH_3CN:0.1 M ammonium formate 3.5:96.5) at a flow rate of 6 ml/min was used to elute the radioligand, which had a retention time of 10 min. The radiochemical purity was greater than 99%. The area of the UV absorbance peak at 254 nm corresponding to carrier product was measured and compared to a standard curve of compound 2 relating mass to UV absorbance.

No-carrier-added radiosynthesis was done under the same reaction conditions. Compound **4** (2 mg) was used as a precursor.

Conclusion

2-[^{18}F]fluoroadenosine **2**, a potential radioligand for assessment of adenylate metabolism, has been synthesized via both carrier-added and no-carrier-added procedures. Further optimization of the radiosynthesis is warranted if this radiotracer proves to be useful in animal studies.

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References

1. Naito K, Ohnishi K. *Gan To Kagaku Ryoho* 2005; **32**: 292–296.
2. Leung WH, Tsang SF, Chim CS. *Am J Hematol* 2005; **79**: 246–247.
3. Zhu Q, Tan DC, Samuel M, Chan ES, Linn YC. *Leuk Lymphoma* 2004; **45**: 2239–2245.
4. Powell AM, Albert S, Oyama N, Sakuma-Oyama Y, Bhogal B, Black MM. *J Eur Acad Dermatol Venereol* 2004; **18**: 360–364.
5. Nabhan C, Gartenhaus RB, Tallman MS. *Leuk Res* 2004; **28**: 429–442.
6. Leporrier M. *Hematol J* 2004; **5** (Suppl 1): S10–S19.
7. Lenz G, Hiddemann W, Dreyling M. *Cancer* 2004; **101**: 883–893.
8. Kowal M, Dmoszynska A, Lewandowski K, Hellmann A, Wegrzyn J, Skotnicki AB, Wolowiec D, Kuliczkowski K, Piszcz J, Kloczko J, Roznowski K, Komarnicki M. *Leuk Lymphom* 2004; **45**: 1159–1165.
9. Johnson SA. *Hematol J* 2004; **5**(Suppl 1): S50–S61.
10. Jackson GH. *Hematol J* 2004; **5**(Suppl 1): S62–S67.
11. Hillmen P. *Hematol J* 2004; **5**(Suppl 1): S76–S86.
12. Boogaerts MA. *Hematol J* 2004; **5**(Suppl 1): S31–S37.
13. Hong JS, Waud WR, Levasseur DN, Townes TM, Wen H, McPherson SA, Moore BA, Bebok Z, Allan PW, Secrist JA, Parker WB, Sorscher EJ. *Cancer Res* 2004; **64**: 6610–6615.
14. Gandhi V, Plunkett W. *Pharmacokinet* 2002; **41**: 93–103.
15. Abraham EH. *PCT Int Appl* WO 2003099342; 2003.
16. Mathews WB, Nakamoto Y, Abraham EH, Scheffel U, Hilton J, Ravert HT, Tatsumi M, Rauseo PA, Traughber BJ, Salikhova AY, Dannals RF, Wahl RL. *Mol Imag Biol* 2005; **7**: 203–208.
17. Montgomery JA, Hewson K. *J Am Chem Soc* 1960; **82**: 463–468.
18. Montgomery JA, Hewson K. *J Am Chem Soc* 1957; **79**: 4559–4562.
19. Krolikiewicz K, Vorbrüggen H. *Nucleos Nucleot Nucl Acids* 1994; **13**: 673–678.

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