III. 5 Chemical Synthesis of 2-Deoxy-2-[\(^{18}\text{F}\)]fluoroacetamido-4-\(\text{O}-(2\text{-amino-2-deoxy-}\beta\text{-D-glucopyranosyl})\)-D-glucopyranose from \([^{18}\text{F}\)]fluoride

*Tada M., Oikawa A., Iwata R.,* Sugiymam H.,** Sato K., Fukuda H.,
Kubota K., Takahashi H., Wakui A., Abe Y., Yamaguchi T.,
Fujiwara T., Sato T., and Ido T.*

Research Institute for Tuberculosis and Cancer, Cyclotron and
Radioisotope Center,* and Chemical Research Institute of Non-
Aqueous Solutions,** Tohoku University

Introduction

In the previous papers,\(^1\sim^3\) we reported that the rapid and efficient one-pot syntheses of 2-deoxy-2-[\(^{18}\text{F}\)]fluoroacetamido-D-glucopyranose (\(N\)-\([^{18}\text{F}\)]fluoroacetyl-D-glucosamine) (1), \(-D\)-mannopyranose, and \(-D\)-galactopyranose, respectively, starting from \([^{18}\text{F}\)]fluoride and ethyl bromoacetate. Sugar (1) has potential as a tumor imaging agent, since mice bearing spontaneous hepatoma showed a high enough concentration of (1) in the tumor for external detection and the rabbit with VX-2 tumor demonstrated clear positron emission tomography (PET) images.\(^4\)

As a part of the synthetic study of sugars labelled with positron emitting nuclides for PET study, this paper describes the rapid one-pot chemical synthesis of 2-deoxy-2-[\(^{18}\text{F}\)]fluoroacetamido-4-\(\text{O}-(2\text{-amino-2-deoxy-}\beta\text{-D-glucopyranosyl})\)-D-glucopyranose (\(N\)-\([^{18}\text{F}\]fluoroacetyl)chitobiose), (2), from \([^{18}\text{F}\)]fluoride and ethyl bromoacetate. Sugar (2) is the first example of oligosaccharide labelled with positron emitting nuclide.

Results and Discussion

Unlabelled 2-deoxy-2-fluoroacetamido-4-\(\text{O}-(2\text{-amino-2-deoxy-}\beta\text{-D-glucopyranosyl})\)-D-glucopyranose (3) was synthesized from 2-amino-2-deoxy-4-\(\text{O}-(2\text{-amino-2-deoxy-}\beta\text{-D-glucopyranosyl})\)-D-glucopyranose (chitobiose), (4), with fluoroacetic acid by the ordinary method using dicyclohexylcarbodiimide (DCC). The yield of (3) based on (4) was 21%. \(N,N'\)-Bisfluoroacetate of (4), (5), was also prepared by the same procedure in a 23% yield.

The characteristic anomeric and C-2 signals of \(^{13}\text{C}\) n.m.r. spectra of (3) and (5) show that the fluoroacetyl group of (3) attaches to the amino group of the reduced end residue. Sugar (3) shows C-2 signals at 55.86 (\(\alpha\)-2), 58.46 (\(\beta\)-2), and 59.17 ppm (\(\beta\)-2') from
internal sodium 2,2,3,3-tetradetero-3-trimethylsilyl-propionate in heavy water and anomeric ones at 92.92 (α-1), 96.98 (β-1), and 105.16 ppm (β-1'). Sugar (5) shows C-2 signals at 55.81 (α-2), 57.92 (β-2') and 58.35 ppm (β-2) and anomeric ones at 92.97 (α-1), 97.04 (β-1), and 103.49 ppm (β-1'). As shown above, the signals of (α and β)-1 and -2 show very similar chemical shifts, but those of β-1' and -2' show different chemical shifts.\(^5,6\)

We recently established the one-pot method for the introduction of a \(^{18}\)Ffluoroacetyl group into aminosugar.\(^1-3\) This method was applied to the synthesis of the title sugar (2) with some modifications.

\(^{18}\)FFluoride was produced by the \(^{18}\)O (p, n) \(^{18}\)F nuclear reaction from a circulating 20% enriched \(^{18}\)O-water target using the Tohoku University Cyclotron.\(^7\) The \(^{18}\)F nuclide thereby formed was converted to potassium \(^{18}\)Ffluoride with potassium carbonate. After addition of 4,7,13,16,21,24-hexaoxa-1,10-diaza bicyclo[8.8.8]hexacosan (Kryptofix 222), the resulting mixture was submitted to the improved one-pot synthesis to give the desired sugar (2) in a 3.7% radiochemical yield. The total synthesis time and radiochemical purity were ca. 80 min and >95%, respectively. The synthetic pathway of (2) from \(^{18}\)Ffluoride and ethyl bromoacetate is shown in Fig. 1. A slight amount of sugar (1), hydrolysis product of (2), was also detected, and the preparative high performance liquid chromatogram (HPLC) is shown in Fig. 2.

Experimental

Kryptofix 222 and plates of thin layer chromatography (TLC) were purchased from E. Merck AG. Ethyl bromoacetate was from Wako Chemical Ltd. and distilled under a reduced pressure. The other reagents were obtained commercially (Wako) and used without further purification. The purity of each compound was always checked by TLC. HPLC analyses were carried out either with a Waters Assoc. (USA) model 6000 equipped with a refractive index detector or with a Waters Assoc. model 4500 equipped with a radioactivity monitor. The packed column [YMC-Pack PA-23 (10.0×250 mm), Yamamura Chem. Lab. Co., Jpn] was used in HPLC. \(^{13}\)C n.m.r. (22.5 MHz) and optical rotation were recorded with a JEOL (Jpn) model FX90Q spectrometer and a JASCO (Jpn) model DIP-181 instrument, respectively. IR spectra were run with a JASCO (Jpn) model A-3 spectrophotometer.

\(N\)-Fluoroacetate (3). A solution of dihydrochloride (413 mg, 1mmol) of (4) in pyridine (1 ml) and water (0.2 ml) was added to a solution of sodium fluoroacetate (120 mg, 1.2 mmol) in 1.5 N hydrochloric acid (1 ml). After addition of DCC (412 mg, 2 mmol) room temperature. The resulting mixture was diluted with water (10 ml) and filtered. The filtrate was washed with ether, concentrated, and passed through an ion retardation resin (AG 11-A8, 2 ml) column using water as elution solvent. The eluent was then concentrated, passed through a Sep-Pak NH\(_{2}\) cartridge, and subjected to preparative HPLC under the
similar conditions in Fig. 2. A peak corresponding to (3) was then collected, and (3) (84 mg) was obtained as colorless oil in a 21% yield. The retention time of (3) is 11.1 min. [α]D

\[ \text{c} \] = 14.6° (c 1.6, water). IR(KBr): 3440 (OH, broad) and 1670 cm\(^{-1}\) (C=O).

\(N,N'-\text{Bisfluoroacetate (5).}\) A mixture of dihydrochloride of (4) (413 mg, 1 mmol) and sodium fluoroacetate (220 mg, 2.2 mmol) was treated as above to give (5) (106 mg) as colorless oil in a 23% yield. The retention time of (5) in HPLC is 9.0 min. [α]D

\[ \text{c} \] = 3.6° (c 2.6, water). IR(KBr): 3430 (OH, broad), 1660 (C=O) and 1630 cm\(^{-1}\) (C=O).

\(2\text{-Deoxy-2-[^{18}\text{F}]fluoroacetamido-4-O-(2-amino-2-deoxy-\beta-D-glucopyranosyl)-D-}

\(\text{glucopyranose (2).}\) [\(^{18}\text{F}\)]Fluoride was produced by the proton bombardment of 20% enriched [\(^{18}\text{O}\)]water.\(^7\) To the aqueous solution of [\(^{18}\text{F}\)]fluoride, a mixture of aqueous potassium carbonate (0.2 ml, 33 μmol) and Kryptofix 222 (27 mg, 72 μmol) was added. The resulting solution was dried at 90°C in a stream of dry nitrogen gas. To the residue, a solution of ethyl bromoacetate (33.4 mg, 0.2 mmol) in acetonitrile (1 ml) was added. The mixture was heated at 82°C for 10 min with stirring and cooled. After addition of 1 N aqueous potassium hydroxide (0.4 ml), the reaction mixture was heated for 5 min, cooled, and neutralized with hydrochloric acid. To the resulting mixture, a mixture of dihydrochloride of (4) (43.2 mg, 0.1 mmol) and DCC (103 mg, 0.5 mmol) in pyridine (0.5 ml) was added. The mixture was treated under similar conditions described in a previous paper 3) to afford a crude reaction product. The product was then submitted to preparative HPLC (Fig. 2). A radioactivity peak corresponding to (2) was then collected, and (2) was obtained in a 3.7% radiochemical yield.

Acknowledgements

The use of the Tohoku University Cyclotron under directions of Professors Manabu Fujioka and Hikonojo Orihara, Tohoku University, is gratefully acknowledged. The present work was supported by Grants-in-Aid for Scientific Research from the Ministry of Education, Science and Culture, Japan.

References

**Fig. 1.** Synthetic pathway of (2) from ethyl bromoacetate and [18F]fluoride.

**Fig. 2.** Preparative HPLC chromatogram of the reaction mixture. A and B are (2) and (1), respectively.