Annals of Nuclear Medicine Vol. 3, No. 3, 143–147, 1989

Preparation of a fine powder of 2-deoxy-2-[^{18}F]fluoro-D-glucose suitable for inhalation to diagnose lung diseases by means of PET

Fumiyoshi OJIMA,* Tatsuo IDO,* Toshihiro TAKAHASHI,* Jun HATAZAWA,* Masatoshi ITO,* Hidetada SASAKI,** Masaru YANAI,** Takashi AIKAWA,** Tamotsu TAKISHIMA,** Kenji ABIKO*** and Masafumi AMANO***

*Cyclotron and Radioisotope Center, Tohoku University
**First Department of Internal Medicine, Tohoku University School of Medicine
***MECT Corporation

Fine 2-deoxy-2-[^{18}F]fluoro-D-glucose (^{18}FDG) powder was obtained by adding diethyl ether into a methyl alcohol solution of ^{18}FDG and other sugar as seed. When micronized particles of sodium N-acetyl-neuraminate (Neu5Ac-Na) were used as seed crystals, particles containing ^{18}FDG were obtained and 80% of them were smaller than 10 μm in size. More than 60% of these crystals were 4–6 μm in size. In a preclinical study of forced inhalation in a dog, the ^{18}FDG fine powder was mainly distributed in the trachea. The radioactivity in the trachea then increased once and a gradual decrease followed. The radioactivity was transferred into the blood and radioactivity incorporation into the heart was observed. After a normal volunteer inhaled ^{18}FDG dry powder aerosol, the radioactivity was found in the respiratory tract and the peripheral area of the lung by means of PET. Absorption and *in vivo* dynamics of the ^{18}FDG were also analysed.

Key words: PET study, ^{18}FDG dry powder aerosol, Mucociliary clearance

INTRODUCTION

IT HAS BEEN RECOGNIZED that in the condition of the mucus on the inner surface of the air tract and clearance of mucociliary transport there are some differences between normals and patients who have chronic bronchitis, bronchial asthma and other lung diseases.[1–3] Quantitative measurement of deposition, clearance of the air tract and absorption from the lung after inhalation of drugs is regarded as useful in diagnosing lung diseases. Noninvasive measurement of the mucociliary clearance by inhalation of ^{99m}Tc particles micronized with a nebulizer has been reported.[4] This method made it possible to analyze the permeability of the alveolar membrane.[5,6] Since the images are planar although the lung is three-dimensional, the details of such regions as the main bronchi and alveoli are not separated. Positron emission tomography (PET) can analyze the deep areas of the whole body exactly and quantitatively and can distinguish the main bronchi and the alveoli. Clearance of the air tract and absorption can be analyzed by periodic measurement. Since ^{18}F has a relatively long half-life among the positron emitting

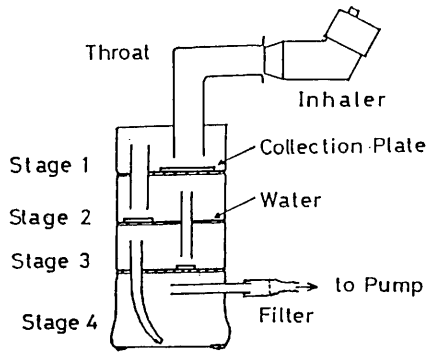


Fig. 1 Diagram showing multistage liquid impinger.

Received June 2, 1989; revision accepted September 11, 1989.

For reprints contact: Tatsuo Ido, Cyclotron and Radioisotope Center, Tohoku University, Aoba, Aramaki, Aoba-ku, Sendai 980, JAPAN.

nuclides, it is suitable for obtaining chronological information. ^{18}FDG is frequently used to measure glucose metabolism in clinical studies. Since ^{18}FDG is soluble in water, it is possibly absorbed from the alveoli into the blood. Therefore, by analyzing the *in vivo* dynamics of ^{18}FDG, some lung diseases may be diagnosed. It is also assumed that analysis of the deposition on the inner surface of the respiratory

Table 1 Effective cut-off diameter for 50% collection efficiency (ECD 50) of multistage liquid impinger

Location	Throat	Stage				Filter
		1	2	3	4	
ECD50 (μm)	27.7	21.2	7.1	5.2	3.3	1.9

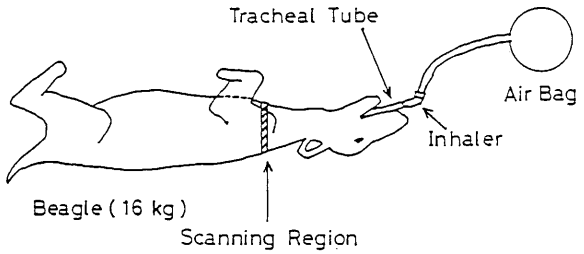


Fig. 2 ^{18}FDG dry powder aerosol administration system for a dog and scanning region.

Table 2 Relationship between radioactivity incorporation yield and methyl alcohol volume

Methyl alcohol (m*l*)	2.0	1.0	0.5	0.25	0.1
Yield (%)	30.7	38.6	45.0	55.1	62.6

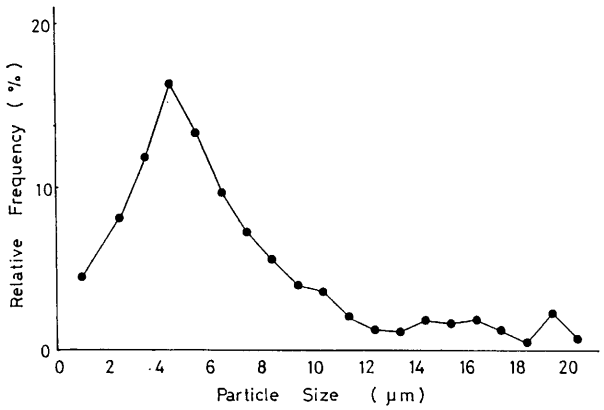


Fig. 3 Particle size distribution of crystals including ^{18}FDG.

Table 3 ^{18}FDG dry powder aerosol dispersion test

Location	Throat	Stage				Filter
		1	2	3	4	
Rate (%)	12.1	34.5	13.1	11.1	8.5	1.3

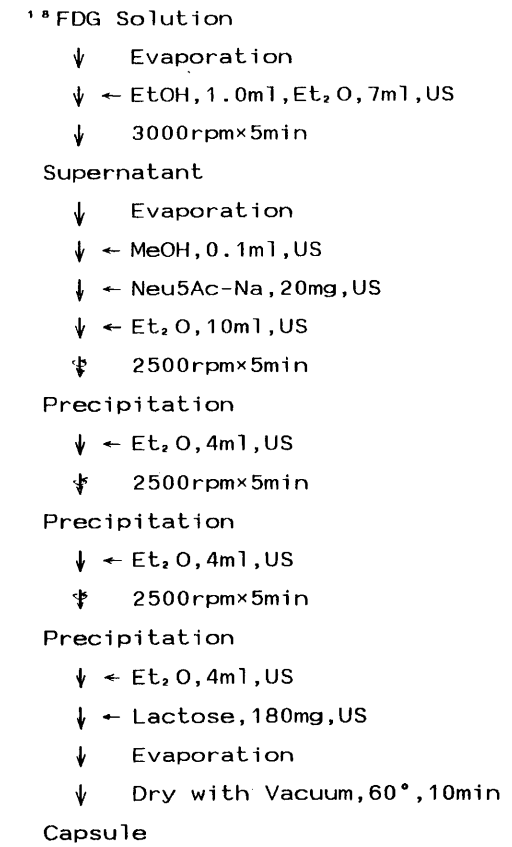


Fig. 4 ^{18}FDG dry powder aerosol preparation procedure. Abbreviations: EtOH=ethyl alcohol; MeOH=methyl alcohol; Et_2 O=diethyl ether; Neu5Ac-Na=sodium N-acetylneuraminate; US=ultrasonication.

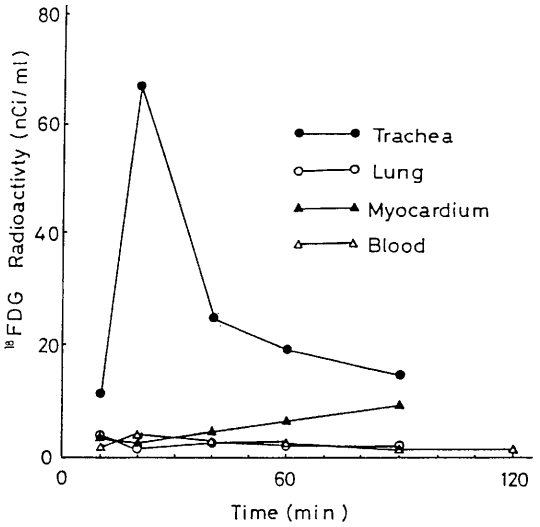
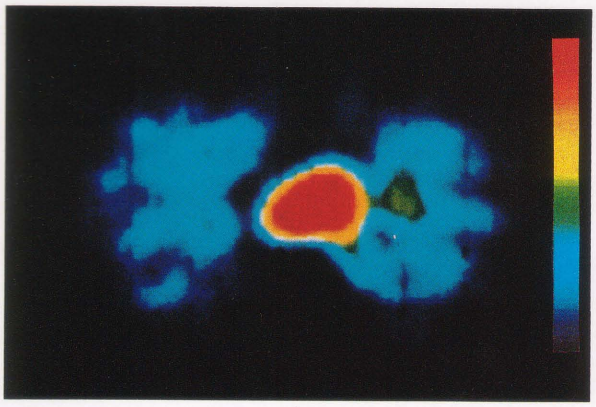
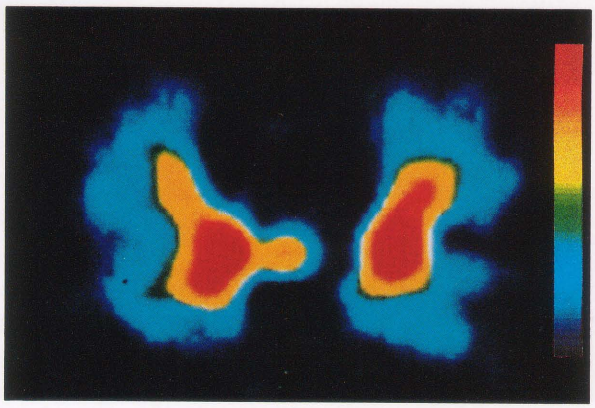


Fig. 5 Elimination and uptake of ^{18}FDG in the trachea, lung, myocardium and blood in the dog.



A



B

Fig. 6 Positron emission tomography of the lung including trachea (A) and mid-heart level (B) after inhalation of ^{18}FDG dry powder aerosol.
Red indicates high radioactivity area and blue indicates low radioactivity area.

Table 4 ^{18}FDG dry powder aerosol dispersion test in normal volunteer

Location	Throat	Stage				Filter
		1	2	3	4	
Rate (%)	7.9	37.6	13.1	10.7	9.3	1.3

tract of ^{18}FDG particles inhaled as a dry powder aerosol will reveal the condition of the mucus. Since drugs in dry powder aerosol form have recently been developed, we examined their availability for administration. In the nebulizer, particle size primarily depends on the apparatus and temperature.[7] However, particles grow larger under such high humidity conditions as in the respiratory tract.[8] The dry powder aerosols seems to be adequate for diagnosis. To examine the usefulness of dry powder aerosols in diagnosis of lung diseases, we investigated the ^{18}FDG fine powder preparation suitable for inhalation.

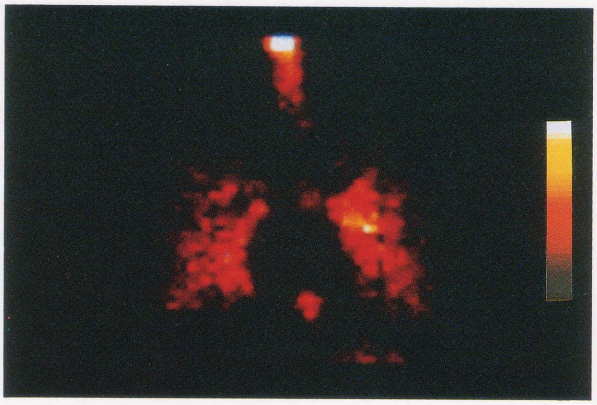


Fig. 7 Rectilinear scan of the lung after inhalation of ^{18}FDG dry powder aerosol.
White indicates high radioactivity area and dark indicates low radioactivity area.

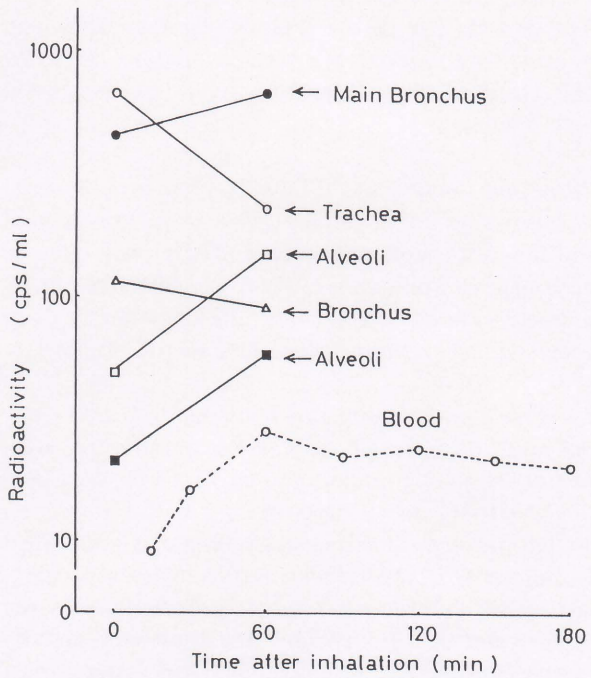


Fig. 8 Change in ^{18}FDG radioactivity in the lung and the blood level after inhalation in human normal volunteer.

MATERIALS AND METHODS

Materials

^{18}FDG was synthesized from triacetylglucal and AcO^{18}F by means of an automated synthesis system.[9] Usually, 5–6 mg of carrier FDG is provided with this system. Sodium-N-acetylneuraminate (Neu5Ac-Na), reported to be nontoxic when inhaled,[10] was obtained from the MECT Corporation (Tokyo). Lactose was purchased from De Melkindustrie Veghel (DMV, Holland) and used after

screening with 105 μm mesh. Other reagents were all special grade.

Preparation of ^{18}FDG *fine particles*

The incorporation yield of radioactivity into crystals and the particle size distribution of seed sugars and crystals including ^{18}FDG were measured in this experiment. The peparation procedure is as follows. Diethyl ether was dropped into a methyl alcohol solution of ^{18}FDG and about 20 mg of sugars (glucose, Neu5Ac-Na and lactose) with ultrasonication (200 W, 38 kHz). Methyl alcohol in the crystals was eliminated by washing twice with diethyl ether and the crystals were mixed with lactose as an additive and dried in a vacuum (60s, 10 min). Approximately 40 mg of the powder was put into hard gelatin capsules for the inhalation experiment.

Particle size distribution analysis

The particle size of the crystals was measured with a centrifugal automatic particle analyzer (CAPA-300, Horiba Ltd.) with n-octyl alcohol as the suspension medium.

Dispersion analysis of ^{18}FDG *dry powder aerosol*

A multistage liquid impinger[11,12] (Fig. 1) was used for the dispersion test of the ^{18}FDG dry powder aerosol at 60 *l*/min air flow for 3 sec. Effective cut-off diameters for 50% collection efficiency (ECD50) in each stage of our apparatus are shown in Table 1.

Preclinical positron emission tomography (PET) study

An adult dog (Beagle, 16 kg) was anesthetized with pentobarbital (25 mg/kg) and 0.47 mCi (17.39 MBq) of the ^{18}FDG powder was inhaled by force with an air bag through intratracheal tubing and a modified Rotahaler (Fig. 2). Inhaled radioactivity was calculated after subtracting residual radioactivity in the inhaler and oral cavity from the total radioactivity in the capsule. Beginning immediately after forced inhalation, positron scans (ECAT II, EG & G, Ortec) and venous blood samples were taken periodically.

In the normal volunteer study, 0.40 mCi (14.8 MBq) of the ^{18}FDG dry powder aerosol was inhaled and two tomographic planes defined by means of X ray CT were imaged for a 46-old-male normal volunteer. One of the slices was an area which included the trachea and the other was primarily the bronchi area at the mid-heart level. Positron scans were done twice at 0 and 60 min. Venous blood samples were taken at 15, 30, 60, 90, 120, 150 and 180 min after inhalation.

RESULTS AND DISCUSSION

Deposition sites of inhaled particles are affected by many factors such as particle size, density, inertia, sedimentation and diffusion. In order to deliver the powder to near the alveoli, the particles must be smaller than about 6 μm.[13] With a pulverizer we could not obtain a powder suitable for inhalation.

Crystallization of ^{18}FDG by adding diethyl ether to the methyl alcohol solution was studied. Crystals of ^{18}FDG could not be obtained by dropping diethyl ether into the methyl alcohol solution without any seeding with sugars. In the sugars employed, smaller particles were obtained when Neu5Ac-Na was used as a seed crystal rather than glucose. Twenty mg of Neu5Ac-Na and ^{18}FDG were dissolved in 2 m*l* of methyl alcohol and then diethyl ether was dropped into this solution at the rate of 10 m*l*/min under ultrasonication. More than 90% of the crystals were smaller than 10 μm, but recovered radioactivity yield in the crystals was not high enough for our inhalation experiment. The relationship between the recovered radioactivity yield and the methyl alcohol volume during crystallization was then investigated (Table 2). When the volume of methyl alcohol was decreased to 0.1 m*l*, the recoved radioactivity yield increased by more than 60% which was sufficient for the experiment. However, Neu5Ac-Na was not completely dissolved in the volume of methyl alcohol used. Since the crystals grew following the deposition of ^{18}FDG on their surface, the particle size was affected by the diameter of the initial Neu5Ac-Na particles. Fig. 3 shows the particle size distribution for 3 experiments with 4–5 μm Neu5Ac-Na as seed. Eighty percent of the crystals were particles smaller than 10 μm. A dispersion test of the ^{18}FDG powder prepared with micronized Neu5Ac-Na was carried out with the multi-stage liquid impinger and the results are shown in Table 3. Thirty percent of the radioactivity dispersed to stages 2, 3 and 4, which were assumed to be the trachea and the bronchi from the ECD50 in Table 1. Examination with fine particles (median diameter, 4–5 μm) of lactose instead of Neu5Ac-Na was carried out, but powder dispersible in the multistage liquid impinger test could not be obtained. Since there was a contamination of an inorganic salt (NaCl) from the ion exchange resin[14] in the synthesized ^{18}FDG, ^{18}FDG was extracted with a mixture of ethyl alcohol and diethyl ether before crystallization. The established preparation procedure for ^{18}FDG dry powder aerosol is shown in Fig. 4. It took about 100 min for the preparation procedure and three capsules of dry powder aerosol including about 2 mCi (74 MBq) of ^{18}FDG were obtained from 50–60 mCi (1.85–2.22 GBq) of the initial ^{18}FDG. Two capsules were

used for the PET study and another capsule was used for the dispersion analysis.

A preclinical study was carried out in a dog with forced inhalation of ^{18}FDG dry powder aerosol. Just after administration of the powder, deposition of the radioactivity in the trachea was recognized. The radioactivity in the trachea increased once and then decreased with time (Fig. 5). The increase possibly reflects the mucociliary clearance of inhaled ^{18}FDG from the deep area of the lung to the measurement site. Since the ^{18}FDG was transferred to an upper site in the throat by mucociliary clearance, a decrease in radioactivity consequently occurred. The ^{18}FDG dry powder aerosol permits measurement of mucociliary clearance in the air tract. Absorption of ^{18}FDG was observed and incorporation into the myocardium was imaged.

In a normal male volunteer, after inhalation of the ^{18}FDG dry powder aerosol it was found that ^{18}FDG was delivered to the trachea, bronchus and peripheral area containing the terminal bronchus and alveoli (Table 4, Figs. 6 and 7). The blood level and elimination of the radioactivity in two planes were measured (Fig. 8). The experiment was performed for up to 60 min and each region was analyzed. The blood level of ^{18}FDG reached its plateau at from 60 min to 180 min. Both the mucociliary transport and absorption from the microcapillaries in the lung could be analysed with this method.

Further studies on lung diseases will be required. The ^{18}FDG dry powder aerosol will be used in diagnosing lung diseases and in revealing the condition of the mucus in the deposition sites in the air tract, since ^{18}FDG is soluble in water.

ACKNOWLEDGMENTS

The authors wish to thank Dr. Koichiro Kawashima for his help in the PET study with the dog, and the members of the Cyclotron and Radioisotope Center for their cooperation.

REFERENCES

1. Hogg JC, Macklem PT, Thurlbeck WM: Site and nature of airway obstruction in chronic obstructive lung disease. *New Engl J Med* 278: 1355–1360, 1968
2. Wanner A: Clinical aspects of mucociliary transport. *Am Rev Respir Dis* 116: 73–125, 1977
3. Goodman RM, Yergin BM, Landa JF, et al: Relationship of smoking history and pulmonary function tests to trachea mucous velocity in nonsmokers, young smokers, ex-smokers, and patients with chronic bronchitis. *Am Rev Respir Dis* 117: 205–214, 1978
4. Isawa T, Teshima T, Hirano T, et al: Lung clearance mechanisms in obstructive airways disease. *J Nucl Med* 25: 447–454, 1984
5. Rees PJ, Shelton D, Chan TB: Effects of histamine on lung permeability in normal and asthmatic subjects. *Thorax* 40: 603–606, 1985
6. Kennedy SM, Elwood RK, Wiggs BJR, et al: Increased airway mucosal permeability of smokers. Relationship to airway reactivity. *Am Rev Respir Dis* 129: 143–148, 1984
7. Pillay M, Akkermans JA, Cox PH: A high efficiency ultrasound nebuliser for radioaerosol studies of the lung. *Eur J Nucl Med* 13: 331–334, 1987
8. Byron PR, Davis SS, Bubb MD, et al: Pharmaceutical implication of particle growth at high relative humidities. *Pestic Sci* 8: 521–526, 1977
9. Iwata R, Ido T, Takahashi T, et al: Automated synthesis system for production of 2-deoxy-2-[^{18}F] fluoro-D-glucose with computer control. *Int J Appl Radiat Isot* 35: 445–454, 1984
10. Takahashi A, Hanabusa T, Mitomi M, et al: Acute inhalation toxicity of a dry powder aerosol sodium-5-(acetoamino)-3,5-dideoxy-D-glycero-D-galacto-2-nonulosonate (KI-111) in rats. *The Clinical Report* 21: 142–146, 1987
11. Bell JH, Hartley PS, Cox JSG: Dry powder aerosols I: A new powder inhalation device. *J Pharm Sci* 60: 1559–1564, 1971
12. May KR: Mutistage liquid impinger. *Bacteriol Rev* 30: 559–570, 1966
13. Landahl HD: On the removal of air-borne droplets by the human respiratory tract: I. The lung. *Bul Math Biophys* 12: 43–56, 1950
14. Reis ML, Draghetta W, Greene LJ: AG 11A8 ion-retardation resin: I. Separation of proteins from sodium chloride, buffer ions, and reagents used to deproteinize solutions. *Anal Biochem* 81: 346–357, 1977