Biological Characters of 6-Deoxy-6-[18F]Fluoro-D-Galactose

III. 3. Biological Characters of 6-Deoxy-6-[^{18}F]Fluoro-D-Galactose

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Introduction

For the assessment of a metabolic function of the liver or tumors in living humans with positron emission tomography (PET), 2-deoxy-2-[^{18}F]fluoro-D-galactose (2-[^{18}F]FdGal) has been proposed as a potential positron-emitting tracer.[1] In the liver and tumor tissues, the tracer is trapped by phosphate and uridylate forms, and incorporation into glycoconjugate macromolecules is substantially inhibited.[2,3] On the other hand, in cell culture study, 6-deoxy-6-fluoro-D-galactose (6-FdGal) is incorporated into cellular glycoconjugate.[4] In this paper we describe *in vivo* characters of 6-[^{18}F]FdGal.

Materials and Methods

Tissue distribution of 6-[^{18}F]FdGal[5] was studied using ddY mice. Two groups of mice were injected intravenously with a mixture of 6-[^{18}F]FdGal and 10 mg D-galactose. For tumor uptake studies, mice with FM3A mammary carcinoma or Lewis lung carcinoma (3LL) and rats with AH109A hepatoma or intracranial KEG-1 glioma were used. The tissue uptake was expressed as the differential absorption ratio (DAR), i.e., (counts of tissue/total injected counts)×(g body weight/g tissue).

The liver, tumor and plasma samples obtained from two FM3A-bearing mice sacrificed 60 min after injection of 6-[^{18}F]FdGal were applied for the analysis of metabolites.[2] Briefly, the samples were treated with $HClO_4$, and were divided into acid-insoluble and acid-soluble fractions. The acid-soluble fraction was analyzed by HPLC.

A rat bearing intracranial KEG-1 tumor was sacrificed 30 min after injection of the tracer. Autoradiography of the brain tumor was performed.[3)]

Results

The results of tissue distribution of 6-[^{18}F]FdGal in mice are summarized in Table 1. Rapid transport of the tracer from blood to all tissues investigated was found; however, radioactivity in these tissues and in blood decreased very rapidly with time except for muscle. At 60 min, 56% of the injected radioactivity was excreted into urine. No effect of D-galactose loading on the liver and brain uptake was shown in Table 2.

All three tumors showed a relatively high uptake and rapid clearance of the radioactivity (Table 3). While tumor-to-blood ratio increased with time, the ratios for other tissues were less than 1.8 and were nearly constant at 2 h. By autoradiography, the higher radioactivity level in KEG-1 brain tumor was visualized compared with the surrounding brain tissue (Fig. 1).

Metabolites of the 6-[^{18}F]FdGal in FM3A bearing mice were summarized in Table 4. Incorporation of small amounts of radioactivity into the acid-soluble fraction, 6-deoxy-6-[^{18}F]fluoro-D-galactonate, 6-[^{18}F]FdGal and UDP-6-[^{18}F]FdGal were identified by comparing their retention times to those of authentic preparations, respectively. Also two unidentified minor metabolites were detected. In liver tissue, galactonate was a main metabolite (39.5%), whereas its ratio in the tumor was only 4%. The galactonate was also found in the plasma.

Discussion

Several *in vitro* studies have shown that 6-FdGal is a biological active analogue of galactose[4,6-8)] However, our in vivo assay does not support that the 6-[^{18}F]FdGal is an applicable tracer for PET studies. Compared to 2-[^{18}F]FdGal,[1-3)] rapid clearance from many tissues, no selective uptake in the liver and tumor and no competition with D-galactose were observed. Small amounts of the 6-[^{18}F]FdGal were certainly incorporated into the macromolecular glycoconjugate via 6-[^{18}F]FdGal-1-P and UDP-6-[^{18}F]FdGal in liver and tumor. However, most radioactivity in liver and tumor was rapidly cleared probably as 6-[^{18}F]FdGal and galactonate forms, because the 6-[^{18}F]FdGal has less affinity for galactokinase[7)] but is oxidized especially in the liver.

Tumor uptake of the tracer was slightly higher compared with several normal tissues. No distinct uptake was found among the three tumors. In the case of 2-[^{18}F]FdGal, two hepatomas showed a slightly higher uptake of the tracer than other tumors[3)]. The brain tumor imaging was demonstrated by autoradiography; however, assessment of the galactose metabolism from the image is difficult.

In conclusion, 6-[^{18}F]FdGal is not suitable as a PET tracer for studying the galactose metabolism *in vivo* in spite of its preferable *in vitro* properties.

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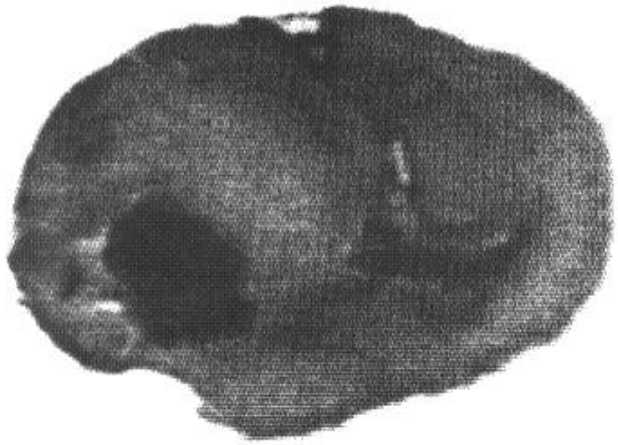

Fig. 1. Autoradiographyc distribution of radioactivity in the coronal section of rat brain with KEG-1 glioma 30 min after injection of 6-deoxy-6-[^{18}F]fluoro-D-galactose.

Table 1. Tissue distribution of radioactivity after i.v. injection of 6-deoxy-6-[^{18}F]fluoro-D-galactose in mice.

	Uptake (DAR)*					
	1 min	5 min	10 min	30 min	60 min	120 min
Blood	1.62±0.25	1.18±0.19	1.05±0.79	0.39±0.09	0.27±0.02	0.13±0.01
Brain	0.89±0.11	0.77±0.08	0.65±0.06	0.26±0.03	0.23±0.03	0.17±0.01
Heart	1.30±0.09	1.04±0.21	0.96±0.02	0.64±0.06	0.60±0.03	0.66±0.08
Lung	1.06±0.19	0.83±0.12	0.82±0.06	0.45±0.09	0.38±0.01	0.24±0.03
Liver	1.83±0.19	1.19±0.21	1.06±0.07	0.55±0.09	0.34±0.02	0.19±0.04
Pancreas	0.52±0.09	0.57±0.14	0.55±0.05	0.37±0.10	0.36±0.09	0.23±0.04
Spleen	0.52±0.09	0.71±0.17	0.75±0.09	0.38±0.04	0.34±0.04	0.24±0.02
Small intestine	1.36±0.07	0.84±0.18	0.75±0.03	0.42±0.04	0.32±0.04	0.21±0.02
Kidney	3.49±0.42	2.00±0.30	1.64±0.21	0.70±0.13	0.55±0.16	0.25±0.02
Muscle	0.65±0.12	0.57±0.11	0.63±0.11	0.51±0.15	0.39±0.08	0.50±0.10

*Mean ±s.d. (n=4)

Table 2. Effect of D-galactose loading on radioactivity levels in the blood, liver and brain after i.v. injection of 6-deoxy-6-[^{18}F]fluoro-D-galactose.

		Uptake (DAR)*	
		10 min	120 min
Liver	Control	1.18±0.13	0.19±0.04
	D-Galactose	1.14±0.10	0.21±0.02
Brain	Control	0.73±0.08	0.17±0.01
	D-Galactose	0.67±0.05	0.18±0.02
Blood	Control	1.24±0.12	0.13±0.01
	D-Galactose	1.22±0.16	0.14±0.02

*Mean ± s.d. (n=4-5)

Table 3. Tumor uptake and tumor-to-tissue uptake ratio after i.v. injection of 6-deoxy-6-[^{18}F]Fluoro-D-galactose in tumor bearing rats and mice.

	Uptake (DAR)*		
	30 min	60 min	120 min
AH109A	0.89±0.06	0.62±0.09	0.34±0.02
FM3A	0.69±0.13	0.39±0.04	0.27±0.07
3LL	0.89±0.23	0.59±0.14	0.38±0.08
	Tumor-to-tissue ratio**		
Blood	1.23 (1.15-1.33)	1.53 (1.05-1.81)	2.48 (1.92-3.17)
Brain	1.68 (1.58-1.87)	1.62 (1.44-1.81)	1.64 (1.29-1.97)
Heart	0.93 (0.71-1.00)	0.74 (0.50-0.92)	0.64 (0.46-0.97)
Lung	1.20 (1.15-1.23)	1.15 (1.02-1.24)	1.28 (0.91-1.73)
Liver	0.94 (0.85-1.00)	0.94 (0.78-1.05)	1.29 (1.06-1.76)
Pancreas	1.80 (1.59-2.11)	1.44 (1.12-1.35)	1.17 (0.96-1.44)
Spleen	1.32 (1.28-1.35)	1.21 (1.11-1.26)	1.17 (0.92-1.24)
Small intestine	1.33 (1.27-1.40)	1.18 (1.07-1.27)	1.27 (0.96-1.44)
Kidney	0.59 (0.49-0.69)	0.65 (0.58-0.69)	0.77 (0.60-0.87)
Muscle	1.73 (1.02-2.55)	1.32 (0.81-1.87)	1.40 (1.03-1.97)

* Mean ± s.d. (n=3-5)
**Mean (range in parenthesis) of the three average ratios calculated in each of three animal models at each time.

Table 4. Metabolites of 2-[^{18}F]fluoro-D-galactose in the plasma, liver and FM3A tumor 60 min after injection.

	Acid-soluble metabolites				Acid-insoluble
	5-[^{18}F]FdGal (%)	Galactonate (%)	Phosphate (%)	Uridylate (%)	materials (%)
Plasma	85.2	12.1	ND	ND	2.7
Liver	50.0	39.5	0.9	2.0	4.5
FM3A	86.3	3.9	*	*	8.4

Data present a mean (n=2).
ND:not detected.
*less than 0.1%.
Retention times of each metabolite were 3.2 min for 6-[^{18}F]FdGal-1-P and 13.2 min for UDP-6-[^{18}F]FdGal.