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Myocardial fatty acid metabolism in diabetic mice with ^{125}I-BMIPP

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In patients with diabetes mellitus, the existence of diabetic cardiomyopathy was substantiated. This study was undertaken to evaluate the myocardiac fatty acid metabolism of diabetic mice (n = 21) and controls (n = 21) in ^{125}I-BMIPP in fasted and unfasted states. ^{125}I-BMIPP of 370 kBq was given and thirty minutes later, animals from both groups were killed. Samples of hearts, liver and other organs were removed, weighed and then counted in a scintillation counter. The percent injected dose/g of hearts of diabetic mice was significantly reduced compared to controls in unfasted ($p < 0.05$) and fasted ($p < 0.01$) groups. These findings may reflect impaired fatty acid utilization of the hearts in diabetic mice compared to controls.

Key words: mouse, diabetes mellitus, fatty acid metabolism, diabetic cardiomyopathy, ^{125}I-BMIPP

INTRODUCTION

IT HAS BEEN REPORTED that diabetes mellitus is associated with an excessive cardiovascular morbidity and mortality.[1] Coronary and peripheral artery atherosclerosis, clinically or at autopsy, is two to three times as prevalent in the diabetic as in the nondiabetic person.[2] Abnormalities of ^{201}Tl, ^{123}I-labeled metaiodobenzylguanidine (MIBG) and glucose metabolism in patients with diabetes mellitus has already been documented.[3–6] Diabetic neuropathy may involve an MIBG abnormality in its early stages.[3] Decreased MIBG uptake in the inferior wall may be a diagnostic sign of cardiac sympathetic dysfucntion in silent myocardial ischemia in diabetes.[4]

^{123}I-labeled 15-(p-idophenyl)-3R,S-methylpentadecanoic acid (^{123}I-BMIPP) is currently widely used as an approved radiopharmaceutical for the evaluation of impairment of myocardial fatty acid metabolism and for myocardial viability.[7–14] Knapp et al. mentioned[7] that this metabolism might make ^{123}I-BMIPP useful in delineating patients with heart disease. The use of ^{123}I-labeled fatty acid may provide complementary information about the metabolic consequences of myocardial viability. Shinmura et al. reported that BMIPP might be used to detect impaired myocardial fatty acid metabolism in patients with diabetes mellitus.[9]

The aim of this study is to examine the myocardial biodistribution of diabetic mice with ^{125}I-BMIPP and also to test the application of BMIPP to diabetic mice for the detection of impaired myocardial fatty acid metabolism.

MATERIALS AND METHODS

Animal models

Fatty acid metabolic studies with ^{125}I-BMIPP were performed at rest in fasting and unfasting states. The study group consisted of male diabetic mice (KK-A^{y}/Ta; NIDDM) and age-matched control mice (C57BL/6J). The mice were 8 wk of age. The body weight of diabetic mice was 38.1 ± 3.75 g and that of the controls was 25.5 ± 1.69 g ($p < 0.01$). The mice were divided into two groups: a fasted group (controls, n = 11; diabetic mouse; n = 11) and an unfasted group (controls, n = 10; diabetic mouse, n = 10). The fasted group were maintained for 24 hr on an overnight fast. The blood glucose level was measured before the ^{125}I-BMIPP study. The blood glucose of the

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Table 1 Biodistribution of ^{125}I-BMIPP in diabetic mice and controls of (Unfasted Group)

(%ID) n = 10

	DM	(Weight)	Controls	(Weight)	p Values
Heart	2.4 ± 0.4	(0.20 ± 0.02)	2.9 ± 1.4	(0.17 ± 0.45)	ns (0.34)
Liver	29.1 ± 6.6		26.6 ± 9.2		ns (0.50)
Lungs	3.8 ± 3.3		5.9 ± 6.4		ns (0.37)
Spleen	0.7 ± 0.2		0.6 ± 0.2		ns (0.53)
Kidneys	3.7 ± 1.1		2.8 ± 0.7		p < 0.05 (0.04)
Digestive Organs	7.1 ± 2.0		6.1 ± 2.1		ns (0.07)
Blood	6.9 ± 5.9		7.2 ± 4.8		ns (0.89)
Others	44.1 ± 6.8		47.4 ± 7.6		ns (0.11)

DM: Diabetic Mellitus

Table 2 Biodistribution of ^{125}I-BMIPP in diabetic mice and controls of (Unfasted Group)

(%ID/g) n = 10

	DM	(Weight)	Controls	(Weight)	p Values
Heart	12.2 ± 2.3	(0.20 ± 0.02)	18.0 ± 8.2	(0.17 ± 0.45)	p < 0.05 (0.04)
Liver	9.4 ± 2.6		15.4 ± 8.0		p < 0.05 (0.04)
Lungs	17.8 ± 12.8		18.4 ± 9.9		ns (0.91)
Spleen	6.8 ± 4.8		6.1 ± 2.4		ns (0.69)
Kidneys	5.3 ± 1.0		6.1 ± 2.1		ns (0.24)
Digestive Organs	5.3 ± 4.8		7.9 ± 2.7		ns (0.36)
Blood	11.0 ± 3.0		12.4 ± 3.5		ns (0.60)
Others	3.0 ± 5.5		1.7 ± 0.8		ns (0.49)

DM: Diabetic Mellitus

Table 3 Biodistribution of ^{125}I-BMIPP in diabetic mice and controls of (Fasted Group)

(%ID) n = 11

	DM	(Weight)	Controls	(Weight)	p Values
Heart	1.2 ± 0.5	(0.16 ± 0.01)	2.7 ± 0.6	(0.12 ± 0.01)	p < 0.01 (0.007)
Liver	19.3 ± 5.1		18.6 ± 2.8		ns (0.70)
Lungs	2.9 ± 5.5		1.1 ± 0.3		ns (0.35)
Spleen	0.5 ± 0.7		0.3 ± 0.1		p < 0.05 (0.04)
Kidneys	3.6 ± 0.7		3.4 ± 0.6		ns (0.43)
Digestive Organs	7.2 ± 2.1		5.9 ± 1.9		ns (0.20)
Blood	11.0 ± 2.3		9.7 ± 2.8		ns (0.38)
Others	53.3 ± 7.1		57.9 ± 4.6		ns (0.08)

DM: Diabetic Mellitus

fasted group was 399 ± 179 mg/d*l* for diabetic mice and 118 ± 19.3 mg/d*l* for controls ($p < 0.01$).

Both groups of mice were intra-peritoneally anaesthetized with pentobarbital sodium (0.2 g/kg) and then 370 kBq of ^{125}I-BMIPP in 0.2 m*l* saline was injected into the caudal vein.

Biodistribution protocol

Mice from both groups were killed at 30 min after the injection. Samples of the hearts and other organs were quickly removed, rinsed with normal saline and weighed. Tracer deposition within the blood, hearts, other organs and carcasses was counted with an animal scintillation counter connected to a multi-channel analyzer. Energy discrimination was proved by a 35% window centered at 35 keV. The tracer activity concentration was expressed as a percentage of the injected dose (%ID) and a percentage of injected dose per gram tissue (%ID/g).

Statistical analysis

All results were expressed as the mean ± s.d. Tracer accumulation in the groups was compared by unpaired t-tests. p Values of less than 0.05 were defined as statistically significant.

RESULTS

The *in vivo* biodistribution of radioactivity following administration of ^{125}I-BMIPP in diabetic mice and controls were demonstrated (Tables 1–4 and Fig. 1). The

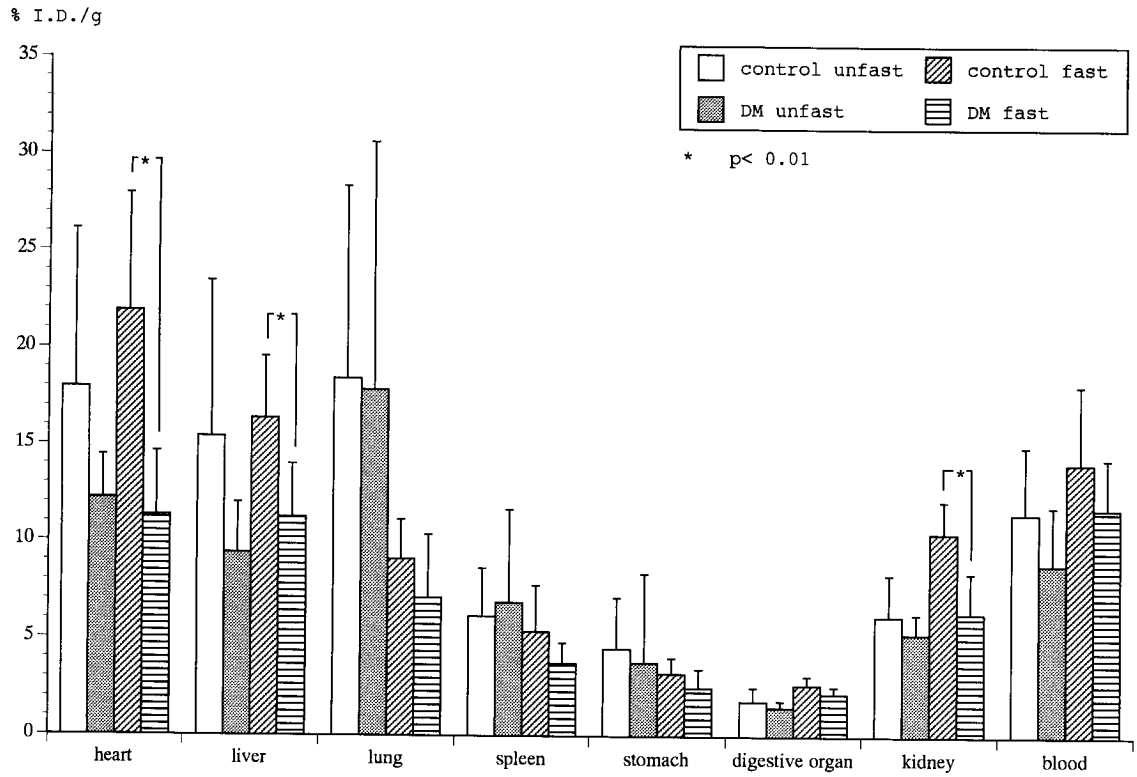

Fig. 1 A comparison of the biodistribution pattern of ^{125}I-BMIPP in diabetic mice and controls at 30 min postinjection. DM: Diabetic Mellitus

Table 4 Biodistribution of ^{125}I-BMIPP in diabetic mice and controls of (Fasted Group)

(%ID/g) n = 11

	DM	(Weight)	Controls	(Weight)	p Values
Heart	11.3 ± 3.3	(0.16 ± 0.01)	22.2 ± 5.6	(0.12 ± 0.01)	$p < 0.01$ (2.2×10^{-5})
Liver	11.2 ± 2.8		16.4 ± 3.2		$p < 0.01$ (0.0007)
Lungs	7.1 ± 3.3		9.1 ± 2.0		ns (0.11)
Spleen	3.7 ± 1.1		5.3 ± 2.4		ns (0.053)
Kidneys	6.2 ± 2.1		10.4 ± 1.7		$p < 0.01$ (4.2×10^{-5})
Digestive Organs	4.5 ± 1.6		5.8 ± 1.3		ns (0.07)
Blood	11.7 ± 2.6		14.1 ± 4.1		ns (0.12)
Others	2.9 ± 0.3		3.0 ± 0.4		ns (0.11)

DM: Diabetic Mellitus

results showed that in the unfasted group the deposition in the hearts (%ID) in diabetic mice and controls was not statistically significant (Table 1), but the %ID/g of the heart of this group was significantly lower in the diabetic mice than in the controls ($p < 0.05$) (Table 2). In contrast, in the fasted group, both the %ID and %ID/g of the hearts were statistically significant between the diabetic mice and controls ($p < 0.01$) (Tables 3 and 4). The %ID of hearts in controls under fasting condition showed the highest activity uptake (Fig. 1). That is, the %ID/g of hearts in controls in the fasted group was almost twice that in the diabetic mice (Fig. 1 and Table 4), but no such great differences were noted in the unfasted group (Tables 1 and 2).

DISCUSSION

^{123}I-BMIPP is not metabolized by beta-oxidation, but is trapped in the triglyceride fraction in the myocardium.[7] Myocardial accumulation of this tracer is associated with triglyceride synthesis, which in part reflects fatty acid utilization. It is also reported that the mitochondrial contribution is predominant in ^{123}I-BMIPP metabolism.[15] The ^{123}I-BMIPP and ^{125}I-BMIPP has been used in clinical and experimental studies in assessing myocardial fatty acid metabolism and viability.[7-14] Abnormal accumulation of ^{123}I-BMIPP in the myocardium has been reported in various diseases and its clinical applicability has also been discussed.[8-10,13]

In patients with hypertrophic cardiomyopathy, ^{123}I-BMIPP imaging may reflect impaired regional fatty acid utilization, which is independent of regional perfusion.[13] On the other hand, diabetes mellitus has been shown to be associated with a specific cardiomyopathy by abundant epidemiologic clinical, and pathologic data.[1] Various pathogenic factors such as small vessel disease, metabolic disease, hypertension, coronary artery disease, autonomic dysfunction, nephropathy and interstitial disease can be explained as pathogenic factors in diabetic cardiomyopathy.[1,16,17] In addition, microvascular morphological abnormalities have been reported in diabetic animals.[18] These microvascular abnormalities may lead to myocardial ischemia in diabetic patients.[17]

In our initial autoradiographic data,[19] we have already revealed diffuse decreased deposit of ^{125}I-BMIPP in the hearts of diabetic mice compared to controls. Shinmura et al.[9] reported that five of the 15 diabetes mellitus patients without coronary artery disease had abnormal BMIPP images.

From the results of the present study, the deposition in hearts (%ID/g) was reduced significantly in the diabetic mice compared to controls under unfasting ($p = 0.04$) and fasting ($p = 2.2 \times 10^{-5}$) conditions (Fig. 1 and Table 4). In controls, the deposition in the hearts was almost twice that in the diabetic mice in the fasted group (Fig. 1 and Table 4), but no such great difference was noted in the unfasted group. These differences shown in our biodistribution studies are thought to be due to the following: the cardiac muscle is capable of utilizing a variety of substrates as sources of energy[20,21]; glucose and fatty acids are the major fuels; also, free fatty acids are the major substrate for energy production in the normal, well-oxygenated heart in the fasted state.[20,21] On the other hand, the reduced myocardial uptake of ^{125}I-BMIPP seen in diabetic mice can be explained as follows: during mild to moderate ischemia, the pattern of substrate uptake changes from a predominant reliance on lipid to a predominantly carbohydrate pattern; free fatty acid uptake is reduced in proportion to the reduction in mitochondrial metabolism.[20,21]

Therefore, in our study in the fasted condition, the control mice utilized fatty acids for major production. As a result, they had a high %ID/g of ^{125}I-BMIPP, but in the diabetic mice with myocardial ischemia,[17] the major energy production is changed from the fatty acid to glucose, so that they had low %ID/g of ^{125}I-BMIPP. On the other hand, in the unfasted condition, the energy production may not be different in the control and diabetic mice compared to under the fasted condition, so that the difference between the control and diabetic mice in %ID/g of ^{125}I-BMIPP became small. It is also noted that the concentrations of plasma substrates, such as blood sugar, free fatty acids, cholesterol, triglyceride and insulin, do not affect the total myocardial uptake, clearance or regional accumulation of ^{123}I-BMIPP.[10] Therefore, ^{123}I-BMIPP can be used in patients with diabetes mellitus to detect impaired fatty acid metabolism.[10]

The results of our study suggest that the metabolism of fatty acid in the heart was impaired in the diabetic mice compared to controls, particularly under fasting conditions. ^{125}I-BMIPP might be useful in investigating the pathogenesis and abnormality of the diabetic heart. In contrast, it has been found that myocardial accumulation of BMIPP decreased compared to flow tracers in various types of disease such as vasospastic angina and hypertrophic cardiomyopathy, etc.[8,12,13] In addition, because it has been reported that the microvascular abnormalities may lead to myocardial ischemia in diabetic patients,[17,18] the examination of fatty acid metabolism with the ^{125}I-BMIPP is not enough to assess the viability of the diabetic myocardium. Further investigation to compare the flow study and ^{125}I-BMIPP in cases of diabetes mellitus would be required.

CONCLUSIONS

The %ID/g of hearts of diabetic mice was significantly reduced compared to controls in unfasted ($p = 0.04$) and fasted ($p = 2.2 \times 10^{-5}$) groups. These findings may reflect impaired fatty acid utilization in diabetic mice compared to controls.

REFERENCES

1. Zarich SW, Nesto RW. Diabetic cardiomyopathy. *Am Heart J* 118: 1000–1012, 1989.
2. Brown AL Jr, Juergens JL. Arteriosclerosis and Atherosclerosis. *In:* Fairbairn II JF, Juergens JL, Spittell JA, eds. *Peripheral Vascular Diseases. fourth ed.* Philadelphia, London, Toronto: W.B. Saunders, pp. 159–171, 1972.
3. Hattori N, Tamaki N, Hayashi T, Masuda I, Kudoh T, Tateno M, et al. Regional abnormality of iodine-123-MIBG in diabetic hearts. *J Nucl Med* 37: 1985–1990, 1996.
4. Matsuo S, Takahashi M, Nakamura Y, Kinoshita M. Evaluation of cardiac sympathetic innervation with iodine-123-metaiodobenzylguanidine imaging in silent myocardial ischemia. *J Nucl Med* 37: 712–717, 1996.
5. Nagamachi S, Jinnouchi S, Nakahara H, Flores LG, Ohnishi T, Hoshi H, et al. ^{123}I-MIBG myocardial scintigraphy in diabetic patients: relationship to autonomic neuropathy. *Nucl Med Commun* 17: 621–632, 1996.
6. Lewis P, Nunan T, Dynes A, Maisey M. The use of low-dose intravenous insulin in clinical myocardial F-18 FDG PET scanning. *Clin Nucl Med* 21: 15–18, 1996.
7. Knapp FF, Ambrose KR, Goodman MM. New radioiodinated methyl-branched fatty acids for cardiac studies. *Eur J Nucl Med* 12: S39–S44, 1986.
8. Nakajima K, Shimizu K, Taki J, Uetani Y, Konishi S, Tonami N, et al. Utility of iodine-123-BMIPP in the diagnosis and follow-up of vasospastic angina. *J Nucl Med* 36: 1934–1940, 1995.
9. Shinmura K, Tani M, Suganuma Y, Hasegawa H, Kawamura M, Nakamura Y, et al. ^{123}I-β-methyl-iodophenyl-pentadecanoic acid myocadial scintigraphy in diabetic patients

without overt ischemic heart disease. *J Cardiol* 26: 23–32, 1995.
10. Takeishi T, Atsumi H, Fujiwara S, Takahashi K, Tomoike H. Fatty acid metabolic imaging with I-123 BMIPP for the diagnosis of coronary artery disease: Application to patients with diabetes mellitus and hyperlipidemia. *Nucl Med Commun* 17: 675–680, 1996.
11. Morishita S, Kusuoka H, Yamamichi Y, Suzuki N, Kurami M, Nishimura T. Kinetics of radioiodinated species in subcellular fractions from rat hearts following administration of iodine-123-labelled 15-(p-iodophenyl)-3-(R,S)-methylpentadecanoic acid (^{123}I-BMIPP). *Eur J Nucl Med* 23: 383–389, 1996.
12. Kurata C, Kobayashi A, Yamazaki N. Dual-tracer autoradiographic study with thallium-201 and radioiodinated fatty acid in cardiomyopathic hamsters. *J Nucl Med* 30: 80–87, 1989.
13. Chen SL, Uehara T, Morozumi T, Yamagami H, Kusuoka H, Nishimura T. Myocardial metabolism of ^{123}I-BMIPP in patients with hypertrophic cardiomyopathy: assessment by radial long-axis SPET. *Nucl Med Commun* 16: 336–343, 1995.
14. Miller DD, Gill JB, Livni E, Elmaleh DR, Aretz T, Boucher CA, et al. Fatty acid analogue accumulation: a maker of myocyte viability in ischemic-reperfused myocardium. *Circ Res* 63: 681–692, 1988.
15. Nitenberg A, Valensi P, Sachs R, Dali M, Aptecar E, Attali JR. Impairment of coronary vascular reserve and ACh-induced coronary vasodilation in diabetic patients with angiographically normal coronary arteries and normal left ventricular systolic function. *Diabetes* 45: 1017–1025, 1993.
16. Nahser PJ, Brown RE, Oskarsson H, Winniford MD, Rossen JD. Maximal coronary flow reserve and metabolic coronary vasodilataion in patients with diabetes mellitus. *Circulation* 91: 635–640, 1995.
17. Tasca C, Stefaneanu L, Vasilescu C. The myocardial microangiopathy in human and experimental diabetes mellitus: a microscopic, ultrastructural, morphometric and computer-assisted symbolic-logic analysis. *Endocrinologie* 24: 59–69, 1986.
18. Oshima M, Higashi S, Kikuchi Y, Fukumitu N, Ban S, Shirai T, et al. Autoradiographic study of myocardial fatty acid metabolism in diabetic mouse using I-125 BMIPP. *Nippon Acta Radiologica* 56: 137–138, 1996.
19. Neely R, Rovetto J, Oram F. Myocardial utilization of carbohydrate and lipids. *Prog Cardiovasc Dis* 15: 289–299, 1972.
20. Opie LH. Fuels: carbohydrates and lipids. *In*: Opie LH, ed. The Heart (*Physiology and Metabolism*). New York: Raven Press, pp. 208–246, 1991.