Evaluation of Myocardial Viability with Iodine-123-BMIPP in a Canine Model

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The tracer ^{123}I-BMIPP was examined for its ability to reflect myocardial lipid metabolism. Studies in mice indicate that myocardial BMIPP uptake correlates with ATP content. Details, however, of myocardial accumulation in the ischemic period with either infarct or ischemia are not well documented. **Methods:** Sixteen adult mongrel dogs were investigated. The occluded left anterior descending artery (LAD) alone was reperfused to make the ischemic area, and the first diagonal branch of the LAD was kept occluded to make the infarct area. Regional wall motion was evaluated by echocardiography in the short-axial view from the epicardium. Tissue blood flow was calculated using nonradioactive colored microspheres. Changes in blood glucose levels, lipid levels and lactate extraction were examined in blood collected from the aorta and great cardiac vein (GCV). The ATP concentration and BMIPP count were determined by high-performance liquid chromatography and gamma-counter, respectively. **Results:** Two hours after reperfusion, blood flow decreased to 20% ± 5% in the infarct area and 64% ± 9% in the ischemic area ($p < 0.05$), despite comparable wall-motion reduction (32% ± 5% and 42% ± 12% in the infarct and ischemic areas, respectively). BMIPP content and ATP concentration showed parallel reduction: 40% ± 7% and 75% ± 4% ($p < 0.05$) of BMIPP and 32% ± 9% and 69% ± 7% ($p < 0.05$) of ATP in the infarct and ischemic areas, respectively. The nonesterified fatty acid extraction, defined as {flow × ([artery] − [GCV])}, decreased to 87% ± 5.6% during occlusion and 75% ± 20.1% 2 hr after reperfusion, as compared with the control value. **Conclusion:** BMIPP uptake correlated well with lipid metabolism and tissue ATP levels and may prove useful in differentiating myocardial infarction from ischemia in the acute phase of ischemic episodes.

Key Words: myocardial viability; iodine-123-BMIPP; lipid metabolism

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The energy metabolic change from beta-oxidation to glycolysis during myocardial ischemia can be evaluated by examining the condition of β-oxidation. Diagnosing ischemia (*1*), and determining its site and the amount of damaged myocardium (*2*) are possible with PET using ^{11}C-palmitate. Distinguishing reduction in blood flow from reduction in fatty acid metabolism can be done by concomitantly using $H_2{}^{15}O$ (*3*). PET machines are few, however, and ^{11}C-palmitate is limited by its tracer which, for instance, allows increased back diffusion from ischemic regions (*4, 5*).

To make noninvasive examination of fatty acid metabolism available at more facilities, fatty acids labeled with various radionuclides have been developed for single-photon emission computed tomography (SPECT). Regional fatty acid metabolism can be evaluated in experimental myocardial ischemia by using ^{123}I-HA (hexadecanoic acid) or ^{123}I-heptadecanoic acid (*6, 7*), which are a straight-chain fatty acids. This technique is useful in differentiating normal from damaged areas and reversible from irreversible myocardial damage (*8, 9*). Furthermore, the uptake and clearance of ^{123}I-IPPA [15-(p-iodophenyl)pentadecanoic acid], a tracer of myocardial lipid metabolism, are useful indices to detect left ventricular wall injuries (*10–13*) and to diagnose ischemia in patients with coronary artery disease (*14*). These straight-chain fatty acids, however, still have problems. The unmetabolized labeled fatty acids diffuse back into circulation without being taken up by myocardial cells, which may cause overestimation of the clearance. In addition, myocardial clearance depends on blood flow, and rapid clearance (*12*) makes myocardial SPECT imaging difficult.

Iodine-123-BMIPP and ^{123}I-DMIPP are ^{123}I-labeled modified fatty acids whose metabolic pathways and clinical application have been evaluated (*15, 16*). In this study the ability of [^{123}I]BMIPP to evaluate myocardial viability was examined in anesthetized and thoracotomized dogs by correlating ^{123}I-BMIPP counts with regional wall motion, regional blood flow, ^{201}Tl counts by dual imaging and myocardial ATP levels.

MATERIALS AND METHODS

Subjects

Sixteen adult mongrel dogs weighing 16.5–30.0 kg were studied. (20 dogs were operated on, but four died of ventricular fibrillation during the experiments despite the use of direct current shock.) Anesthesia was induced by intramuscular injection of Ketaral at 2.5 mg/kg and maintained by intravenous infusion of nembutal at 25 mg/kg. After endotracheal intubation, the animals were connected to a respirator supplying 100% oxygen at 2 liter/min. Catheters were inserted into the bilateral femoral arteries: one was connected to a polygraph for blood pressure monitoring; the other was used for blood sampling and blood flow measurement. A triple-lumen intravenous catheter was placed in the femoral vein for fluid supplementation and drug administration. Thoracotomy was performed at the fifth intercostal space, and the pericardium was fixed to the thoracic wall in a cradle. A catheter was inserted into the left atrial appendage for infusion of colored microspheres, [^{123}I]BMIPP, and Evans blue dye before killing. As shown in Figure 1, an occluder and a Doppler flow meter were connected to the left anterior descending artery (LAD) at its proximal portion, and an occluder was attached also to the first diagonal branch (D). Electrocardiograms were monitored by limb leads and recorded with the blood pressure, dP/dT, and coronary blood flow (Doppler velocity) using a polygraph. All grossly observable collaterals were ligated on the epicardial surface. Blood was drawn from the great cardiac vein (GCV) and LAD artery before occlusion (control), 30 min after occlusion, and 20 and 120 min after reperfusion of the LAD, and changes in myocardial substrates and metabolites were examined. Blood glucose (BS), lactate (LAC), nonesterified fatty acids (NEFA), triglycerides (TG), phospholipids (PL) and total

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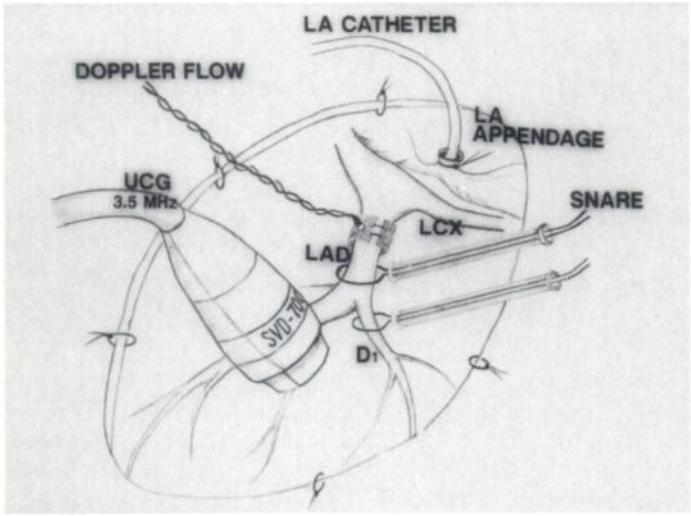

FIGURE 1. Illustration of experimental dog. The dog chest was opened and heart was kept in a cradle. UCG probe was attached to epicardium to calculate regional wall motion. Doppler flow probe was equipped just proximal to first diagonal branch. The occluder (snare) was set both at left anterior descending artery (LAD) and first diagonal branch (D1). Left atrial (LA) catheter was inserted through LA appendage. LA = left atrium; LAD = left anterior descending artery; LCX = left circumflex artery; D1 = first diagonal branch; UCG = ultrasonic cardiography.

cholesterol (T-Chol) were measured. Both occluders were closed for 20 sec, and the magnitude of the resulting reactive hyperemia was determined. The protocol was started when coronary blood flow returned to control level. At the end of the protocol, Evans blue dye was infused to determine the ischemic area, and the animals were killed. The heart was promptly excised and cut into 3–4 slices along the short axis; myocardial sections were collected from the epicardium and endocardium of the LAD, D and normal regions for ATP assay. Slices were stained with triphenyl tetrazorium chloride (TTC) to determine the infarct area, then imaged ex vivo. Ten pieces each from the LAD, D and normal regions were collected again as for the ATP assay, and [^{123}I]BMIPP and ^{201}Tl uptakes were counted. (The ^{201}Tl uptake was counted again after 48–72 hr, when ^{123}I had almost disappeared.) Each piece was used later for tissue blood flow measurement.

Experimental Protocol

First, both the LAD and D were occluded. Thirty minutes later, the LAD was selectively reperfused and the D continuously occluded. After 2 hr reperfusion, ^{123}I-15-ρ-iodophenyl-3-(R,S)-methylpentadecanoic acid ([^{123}I]BMIPP) and ^{201}Tl, which were kindly given by Nihon Medi-physics Co., Ltd., Japan, were infused at 111 MBq each into the left atrial appendage, and in vivo imaging was performed.

Regional wall motion of the myocardium was evaluated by echocardiography before occlusion (control) and 20, 60, 90 and 120 min after occlusion (before infusion of ^{123}I-BMIPP). The probe was directly applied to the epicardium; short-axis images at the papillary muscle level were recorded for the D region, and short-axis images from the papillary muscle level to the apex were recorded for the LAD region. The endocardium was traced in the end-diastolic and end-systolic images, and the traces were analyzed by the centerline method. Also evaluated were the mean value in the index region, calculated as the mean − 2 s.d. or less of the value determined in occluded control dogs, and its changes with time as percent of the control value (100%) (*17*).

Absolute myocardial blood flow was examined with nonradioactive colored microspheres before (control), 20 min and 120 min after coronary occlusion. Colored microspheres were thoroughly suspended using two syringes connected by a three-way stopper and infused in a bolus at 200,000 microspheres/kg from the left atrial appendage. Simultaneously, blood was aspirated from the femoral artery at 10 ml/min for 90 sec. Tissue specimens were obtained from the 3–4 short-axial slices of the heart excised after death. Two pieces (0.5–1.0 g/piece) each from the epicardium and endocardium of the LAD, D and normal regions (12 pieces altogether) were subjected to Evans blue and TTC staining. After the pieces were repeatedly treated with blood and tissue lysing reagents and centrifuged, the regional myocardial blood flow values were calculated by a formula using the microscopic counts (*18*).

To determine the ischemic area, a 1.0% Evans blue solution was infused from the left atrial appendage immediately before death; to determine the infarct area, slices were subjected to TTC staining and non-stained areas were traced. Ischemic and infarct areas were calculated as a percentage of the whole cut surface on both sides of each slice, and were averaged and multiplied by slice weight. All the ischemic and infarct areas were summed up and expressed as a percentage of the left ventricular (LV) area.

For the ATP assay, the excised tissue was weighed and promptly frozen in liquid nitrogen. The tissue was crushed in a mortar with 3 ml of 0.42 *M* perchloric acid. The tissue homogenate was centrifuged at 4°C and 3000 rpm for 10 min. After neutralization with 6 N potassium hydroxide, 2 ml of the supernatant was centrifuged at 4°C and 3000 rpm for 10 min, and the supernatant was stored at −20°C. ATP, ADP, and AMP were identified and assayed by high-performance liquid chromatography (*19, 20*).

The [^{123}I]BMIPP and ^{201}Tl uptake counts, which were determined after the ^{123}I counts decreased, were expressed as the percentage of the uptake counts in the LAD and D regions relative to those in the normal region.

Data Analysis and Statistics

The mean and s.d. values were calculated from the data obtained; their differences were examined by Student's t-test, and their changes with time were examined by ANOVA at the $p < 0.05$ significance level. Data in the figures are expressed as the mean ± s.e.

RESULTS

Table 1 shows changes in blood pressure, heart rate and double products throughout the study. The heart rate was

TABLE 1
Double Products, Blood Pressure and Heart Rate Measurements

	Cont	Occ30min	Rep20min	Rep60min	Rep90min	Rep120min
DP × 10^3	232 ± 17	209 ± 14*	192 ± 15†	194 ± 15†	205 ± 19	207 ± 18*
BP (mmHg)	146 ± 6	140 ± 6	134 ± 6*	135 ± 6*	140 ± 8	143 ± 7
HR (beat/min)	157 ± 5	147 ± 4†	141 ± 5†	141 ± 4†	142 ± 5†	142 ± 5†

*$p < 0.05$ vs. control.
†$p < 0.01$ vs. control.
Cont = control; Occ = occlusion; Rep = reperfusion; DP = double products; BP = blood pressure; HR = heart rate.

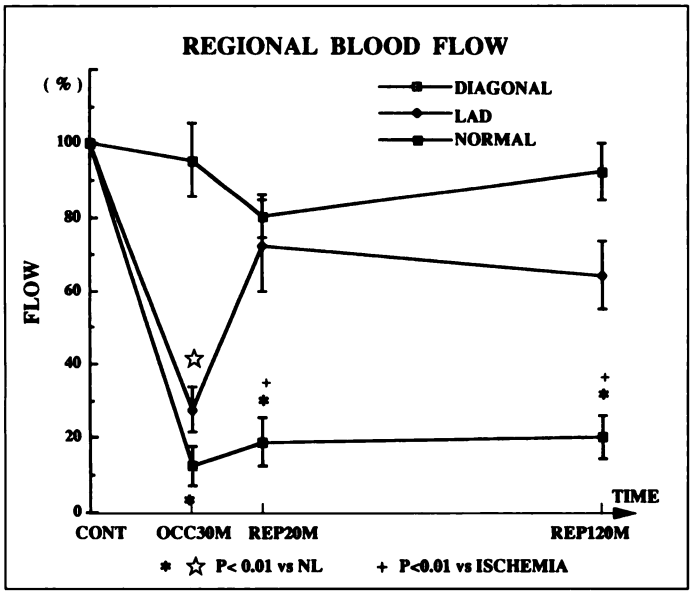

FIGURE 2. Change in regional myocardial flow is shown. Although diagonal and LAD flow decreased significantly after 30 min of occlusion (OCC 30), LAD flow recovered well compared to diagonal flow after reperfusion. Flow at normal area showed no significant difference through the experiment. CONT = control; OCC = occlusion; REP = reperfusion; M = minutes.

significantly decreased 30 min after occlusion but was stable thereafter until the end of the experiment. As shown in Figure 2, regional blood flow, calculated by colored microspheres, decreased to 30% or less immediately after occlusion in both the LAD and D regions (27.5% ± 6% and 12.6% ± 5%, respectively). Two hours after reperfusion, blood flow increased in the LAD region but remained significantly lower in the D region compared to that in the LAD (64% ± 9% and 20% ± 5%, respectively).

The ischemic area estimated by Evans blue staining was 31.6% ± 2.1%, and the infarct area estimated by TTC staining was 4.9% ± 0.9% of the LV weight. Regional wall motion (Fig. 3) did not significantly differ between the LAD and D regions during occlusion or after reperfusion. Two hours after reperfusion of the LAD region, it fell to 42% ± 12% and 32% ± 5% in LAD and D, respectively.

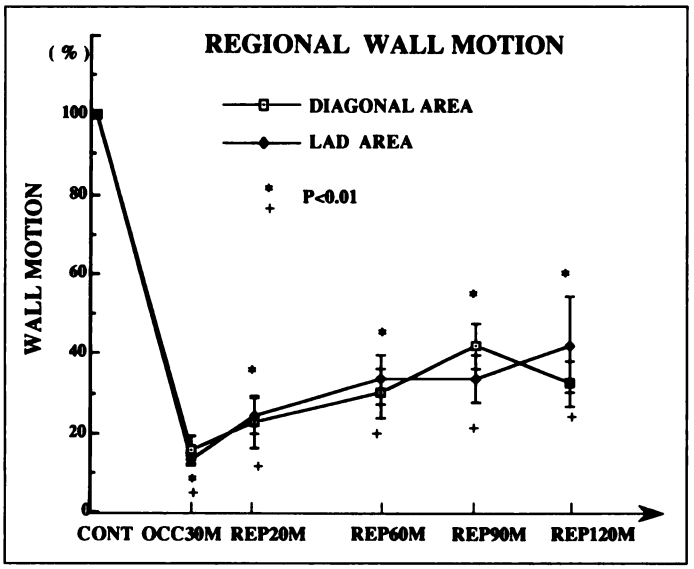

FIGURE 3. Change in regional wall motion is shown. Regional wall motion at both LAD and diagonal area showed significant decrease after 30 min of occlusion (OCC 30), and both region showed no significant difference in recovery of wall motion after reperfusion. Abbreviations are the same as in Figure 2.

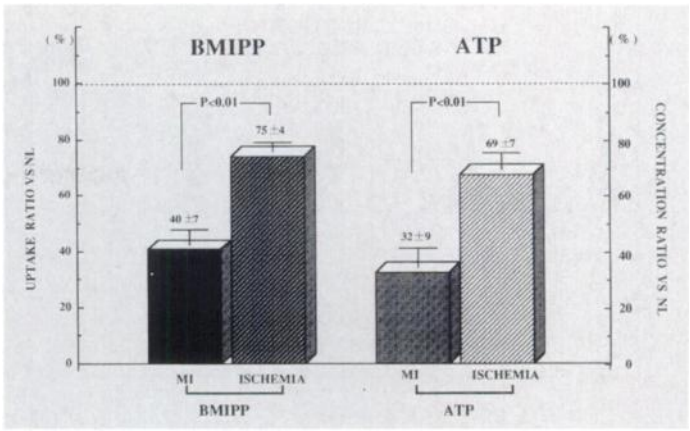

FIGURE 4. Uptake ratio of ^{123}I-BMIPP and ATP content compared to normal region is shown. BMIPP and ATP content both at infarct (MI:diagonal) and ischemic area (ISCHEMIA:LAD) showed comparable change after the procedure. ATP content was slightly lower in relative uptake than BMIPP at both area. NL = normal.

Figure 4 depicts ^{123}I-BMIPP uptake and ATP levels in myocardial samples. The ^{123}I-BMIPP uptake was 75% ± 4% of control in the LAD region and 40% ± 7% of control in the D region, with a significant difference ($p < 0.01$). Similarly, ATP concentrations decreased to 69% ± 7% and 32% ± 9%, respectively. An ex vivo image of BMIPP is shown in Figure 5, and in vivo imaging qualitatively agreed with the tracer's counts. The monitored in vivo image also reflected the ex vivo image, although quantitative analysis was not done.

Figure 6 shows changes in myocardial energy substrate metabolism, with values representing (blood flow) × [arterial blood concentration − GCV blood concentration]. Glucose utilization and lactate production rose, and free fatty acid utilization fell (87% ± 5.6% during occlusion and 75% ± 20.1% 2 hr after reperfusion), compared with the control. The decreased free fatty acid utilization and increased lactate pro-

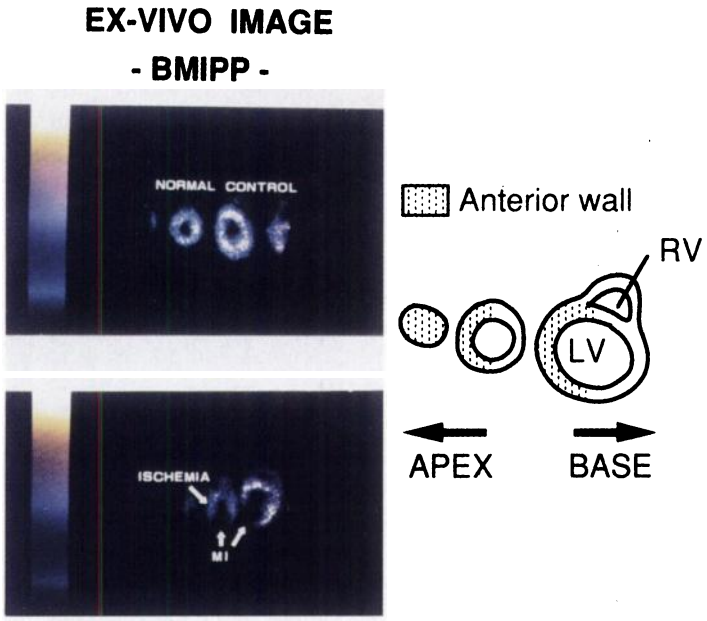

FIGURE 5. Ex vivo image of BMIPP using four slices from apex to base. The image of normal dog (upper) shows diffuse myocardial uptake of BMIPP, however, an ischemic dog showed total defect at infarct region (diagonal area), and decreased uptake at ischemic region (distal left anterior descending area).

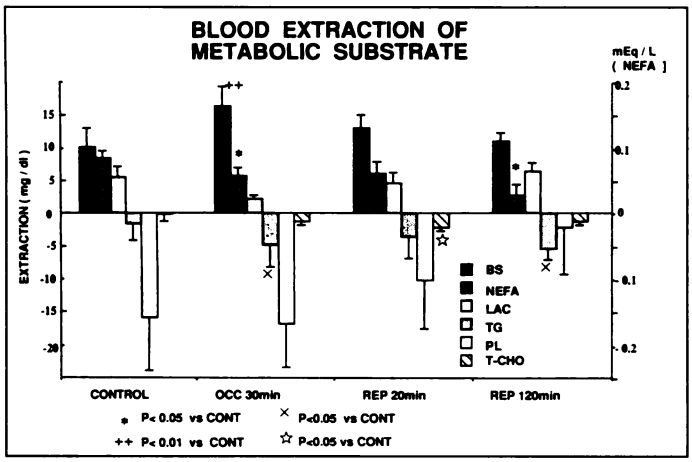

FIGURE 6. Change in myocardial extraction of energy substrates including lipid. Symbols are the same as in Figure 2. BS = blood sugar; NEFA = nonesterified fatty acid; LAC = lactate; TG = triglyceride; PL = phospholipid; T chol = total cholesterol.

duction persisted until 20 and 120 min after LAD reperfusion. Comparison of [^{123}I]BMIPP and ^{201}Tl uptake revealed some differences between the reperfused (LAD) and infarct (D) regions. In the LAD region, most samples (87.5%) showed higher uptake of ^{201}Tl than ^{123}I-BMIPP [correlation coefficient between [^{123}I]BMIPP (X) and ^{201}Tl (Y) was r = 0.96; the regression line Y = 0.94x + 0.084]. In the D region, however, all but one sample showed lower uptake of ^{201}Tl than [^{123}I]BMIPP (r = 0.98; Y = 0.82x + 0.016).

DISCUSSION

In this study, using anesthetized and thoracotomized dogs, we evaluated changes in [^{123}I]BMIPP uptake resulting from myocardial ischemia or myocardial injury and the relationship of such changes with myocardial viability. The protocol included a 30-min occlusion of the LAD followed by reperfusion with sustained occlusion of the diagonal branch to produce regions clearly different in ischemia severity. Wall motion, blood flow and ATP levels were measured in each region. ^{123}I-BMIPP count was found to be significantly related to ischemia severity and myocardial viability but not to wall motion.

Iodine-123-BMIPP, a tracer developed for examination of myocardial fatty acid metabolism, has a methyl branch in the beta position and is therefore resistant to beta-oxidation, which results in delayed myocardial clearance, long retention and high accumulation in the myocardium (*15, 16*). Because of low liver uptake and rapid excretion from the liver, [^{123}I]BMIPP provides clear cardiac images and information useful for evaluating the mechanism of myocardial metabolism (*21, 22*). Iodine-123-BMIPP yields different data in myocardial infarction and angina pectoris (*23*) and has an accumulation pattern different from ^{201}Tl (*24*).

Yonekura et al. (*25*) and Kurata et al. (*26*) reported a "mismatch" between [^{123}I]BMIPP and ^{201}Tl uptake in hypertensive rats and cardiomyopathic hamsters. They found that defects were larger with ^{201}Tl than with [^{123}I]BMIPP, that the ^{123}I-BMIPP count had regional heterogeneity, and therefore that [^{123}I]BMIPP is more useful than ^{201}Tl in evaluating cardiomyopathy or drug effects. In comparing [^{123}I]BMIPP with ^{201}Tl in patients, Yamamoto et al. (*24*) observed that the distribution of the two tracers differed in normotensive and hypertensive individuals and that [^{123}I]BMIPP was distributed heterogeneously. Reduced endocardial uptake was especially pronounced in cases of prolonged severe hypertension, suggesting that BMIPP may be helpful in elucidating the role of mitochondrial change in the etiology of hypertrophy.

When Fujibayashi et al. (*27*) treated mouse myocardia with a dinitrophenyl electron transport uncoupler (DNP), they found reduced myocardial [^{123}I]BMIPP uptake which correlated with diminished ATP levels. In the ischemic area, we observed blood flow recovery after reperfusion, but there were slight decreases in the ATP level and the [^{123}I]BMIPP uptake relative to the normal region. Therefore, [^{123}I]BMIPP uptake is considered to reflect myocardial ATP levels during ischemia. Recently, we reported that BMIPP uptake sensitively reflects mitochondrial function in acute ischemia (*28*). Regional wall motion of the myocardium was reduced in the LAD and D regions to the same degree, even after reperfusion and regardless of the degree of ischemia, and sustained ^{123}I-BMIPP uptake is supposed to reflect myocardial viability. Moreover, preserved oxidative metabolism in mitochondria, suggesting ATP production, is required for survival of ischemically injured myocardium (*29*). Dean et al. reported that the injured myocardium maintains a paradoxically high oxygen consumption and that, despite decreased contractility and abnormal wall motion, mitochondrial function was normal after 15-min coronary artery occlusion and reperfusion (*30*). Our findings indicate that preserved mitochondrial function with preserved ATP and oxidative metabolism is a good marker of myocardial viability.

Imaging with the BMIPP tracer is particularly useful because of its slow clearance time. In addition, our study reveals that extraction and retention remain as high as 70%–90% of the arterial content, using an Etomoxir mitochondrial carnitine shuttle blocker or after 10-min occlusion-reperfusion in ischemia. Some back diffusion occurred, however, depending on ischemia severity (*28, 31*).

The ^{201}Tl count was lower than the [^{123}I]BMIPP uptake in the D region, in which severe ischemia was sustained or an infarct was made, but higher in the reperfused ischemic area. The differences in uptake counts were not large, however, and may be compatible with the differences of defect area found in human ^{201}Tl and [^{123}I]BMIPP imaging. Dual imaging with [^{123}I]BMIPP and ^{201}Tl allows determination of the area of ischemia-induced metabolic disturbance and, as has been indicated by clinical reports (*23*) provides more information about myocardial injury than either tracer alone. Mismatch of their images may predict future recovery of the injured myocardium (*32*).

Although ATP and [^{123}I]BMIPP were decreased to a similar degree in the ischemic as compared with the normal region (Fig. 4), the [^{123}I]BMIPP counts were determined in tissue sections adjacent to those used for measurement of ATP, which were collected immediately after death. The relationship between them, therefore, cannot be evaluated by direct comparison. Nevertheless, the reductions were parallel in each animal.

CONCLUSION

The [^{123}I]BMIPP counts correlated well with tissue ATP levels and were related inversely to the severity of blood flow disturbance. Therefore, [^{123}I]BMIPP imaging should be useful for differential diagnosis between myocardial infarction and transient myocardial ischemia during acute ischemia. Finally, [^{123}I]BMIPP imaging allowed successful evaluation of myocardial viability despite similar reductions in wall motion.

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REFERENCES

1. Weiss ES, Hoffman EJ, Phelps ME, et al. External detection and visualization of myocardial ischemia with ^{11}C-substrates in vitro and in vivo. *Circulation* 1976;39: 24–32.
2. Ter-Pogossian MM, Klein MS, Markham J, Robert R, Sobel BE. Regional assessment of myocardial metabolic integrity in vivo by positron-emission tomography with ^{11}C-labeled palmitate. *Circulation* 1980;61:242–255.
3. Lerch RA, Bergmann SR, Ambos HD, Welch MJ, Ter-Pogossian MM, Sobel BE. Effect of flow-independent reduction of metabolism on regional myocardial clearance of ^{11}C-palmitate. *Circulation* 1982;65:731–738.
4. Fox KAA, Abendschein DR, Ambos HD, Sobel BE, Bergmann SR. Efflux of metabolized and nonmetabolized fatty acid from canine myocardium. *Circ Res* 1985;57:232–243.
5. Rosamond TL, Abendschein DR, Sobel BE, Bergmann SR, Fox KAA. Metabolic fate of radiolabeled palmitate in ischemic canine myocardium: implications for positron emission tomography. *J Nucl Med* 1987;28:1322–1329.
6. Van der Wall EE, Westera G, den Hollander W, Visser FC. External detection of regional myocardial metabolism with radioiodinated hexadecanoic acid in the dog heart. *Eur J Nucl Med* 1981;6:147–151.
7. Schon HR, Senekowitsch R, Berg D, et al. Measurement of myocardial fatty acid metabolism: kinetics of iodine-123 heptadecanoic acid in normal dog hearts. *J Nucl Med* 1986;27:1449–1455.
8. Van der Wall EE, Heidendal GAK, den Hollander W, Westera G, Roos JP. Iodine-123-labeled hexadecanoic acid in comparison with thallium-201 for myocardial imaging in coronary heart disease. *Eur J Nucl Med* 1980;5:401–405.
9. Van der Wall EE, Heidendal GAK, den Hollander W, Westera G, Roos JP. Metabolic myocardial imaging with ^{123}I-labeled heptadecanoic acid in patients with angina pectoris. *Eur J Nucl Med* 1981;6:391–396.
10. Van der Wall EE, Den Hollander W, Heidendal GAK, Westera G, Majid PA, Roos JP. Dynamic myocardial scintigraphy with ^{123}I-labeled heptadecanoic acid in patients with angina pectoris. *Eur J Nucl Med* 1981;6:383–389.
11. Lenzhofer R, Dudczak R. Indication of doxorubicin cardiotoxicity by impairment of ^{131}I-I-pIPPA utilization. *Eur J Nucl Med* 1986;12:S32–S33.
12. Reske SN, Sauer W, Machulla HJ, Knust J, Winkler C. Metabolism of 15(p-^{123}I-iodophenyl) pentadecanoic acid in heart muscle and noncardiac tissues. *Eur J Nucl Med* 1985;10:228–234.
13. Rellas JS, Corbett LR, Kurkarni P, et al. Iodine-123 phenylpentadecanoic acid: detection of acute myocardial infarction and injury in dogs using an iodinated fatty acid and single-photon emission tomography. *Am J Cardiol* 1983;52:1326–1332.
14. Kennedy PL, Corbett JR, Kulkarni PV, et al. Iodine ^{123}I-phenylpentadecanoic acid myocardial scintigraphy: usefulness in the identification of myocardial ischemia. *Circulation* 1986;74:1007–1015.
15. Knapp FF, Goodman MM, Callahan AP, Kirsch G. Radioiodinated 15-(p-Iodophenyl)-3, 3-dimethylpentadecanoic acid: a useful new agent to evaluate myocardial fatty acid uptake. *J Nucl Med* 1986;27:521–531.
16. Knapp FF, Ambrose KR, Goodman MM. New radioiodinated methyl-branched fatty acids for cardiac studies. *Eur J Nucl Med* 1986;12:S39–S44.
17. Okuda K, Nohara R, Fujita M, Tamaki N, Konishi J, Sasayama S. Technetium-99m pyrophosphate uptake as an indicator of myocardial injury without infarct. *J Nucl Med* 1994;35:1366–1370.
18. Hale SL, Alker KJ, Kloner RA. Evaluation of nonradioactive, colored microspheres for measurement of regional myocardial blood flow in dogs. *Circulation* 1988;78:428–434.
19. Sellevold OFM, Jynge P, Aarstad K. High-performance liquid chromatography: a rapid isocratic method for determination of creatine compounds & adenine nucleotides in myocardial tissue. *Mol Cell Cardiol* 1986;18:517–527.
20. Takeyama N, Yakagi D, Adachi K, Tanaka T. Measurement of free and esterified carnitine in tissue extracts by high-performance liquid chromatography. *Anal Biochem* 1986;158:346–354.
21. Dudczak R, Schmoliner R, Angelberger P, Knapp FF, Goodman MM. Structurally modified fatty acids: clinical potential as tracers of metabolism. *Eur J Nucl Med* 1986;12:S45–S48.
22. Ambrose KR, Owen BA, Goodman MM, Knapp FF. Evaluation of metabolism in rat hearts of two new radioiodonated 3-methyl-branched fatty acid myocardial imaging agents. *Eur J Nucl Med* 1987;12:486–491.
23. Nishimura T, Sago M, Kihara K, et al. Fatty acid myocardial imaging using ^{123}I-β-methyl-iodophenyl pentadecanoic acid (BMIPP): comparison of myocardial perfusion and fatty utilization in canine myocardial infarction (occlusion and reperfusion model). *Eur J Nucl Med* 1989;15:341–345.
24. Yamamoto K, Som P, Brill B, et al. Dual tracer autoradiographic study of β methyl-(1-^{14}C) heptadecanoic acid and 15-p-(^{131}I-iodophenyl)-β-methylpentadecanoic acid in normotensive and hypertensive rats. *J Nucl Med* 1986;27:1178–1183.
25. Yonekura Y, Brill AB, Som P. Quantitative autoradiographic measurements of regional myocardial substrate utilization in hypertensive rats. *Science* 1985;227:1494–1496.
26. Kurata C, Kobayashi A, Yamazaki N. Dual tracer autoradiographic study with thallium-201 and radioiodinated fatty acids in cardiomyopathic hamsters. *J Nucl Med* 1989;30:80–87.
27. Fujibayashi Y, Yonekura Y, Takemura Y, et al. Myocardial accumulation of iodinated β-methyl-branched fatty acid analogue, iodine-125-15-(p-iodophenyl)-3-(R,S)-methylpentadecanoic acid (BMIPP), in relation to ATP concentration. *J Nucl Med* 1990;31:1818–1822.
28. Hosokawa R, Nohara R, Okuda K, et al. Cardiac metabolism of ^{123}I-BMIPP: comparison with coronary normal dogs, those pretreated with Etomoxir and those with ischemia [Abstract]. *J Nucl Cardiol* 1995;2(suppl):S38.
29. Czermin J, Porenta G, Brunken R, et al. Regional blood flow, oxidative metabolism and glucose utilization in patients with recent myocardial infarction. *Circulation* 1993;88:884–895.
30. Dean EN, Shlafer M, Nicklas JM. The oxygen consumption paradox of "stunned myocardium" in dogs. *Basic Res Cardiol* 1990;85:120–131.
31. Hosokawa R, Nohara R, Okuda K, et al. Cardiac metabolism of I-123-β-methyl-p-iodophenyl pentadecanoic acid (I-123 BMIPP) in normal dogs and those treated with carnitine palmitoyl-transferare, inhibitor (Etomoxir): contribution of α- and β-oxidation [Abstract]. *J Nucl Med* 1995;36(suppl):137P.
32. Tamaki N, Kawamoto M, Yonekura Y, et al. Regional metabolic abnormality in relation to perfusion and wall motion in patients with myocardial infarction: assessment with emission tomography using an ionated branched fatty acids. *J Nucl Med* 1992;33:659–667.