^{99m}Tc-Tetrofosmin Assessment of Myocardial Perfusion and Viability in Canine Models of Coronary Occlusion and Reperfusion

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The goal of this study was to determine whether ^{99m}Tc-tetrofosmin can assess regional flow heterogeneity when injected during sustained coronary artery occlusion and to estimate the degree of myocardial salvage and viability during coronary reperfusion. **Methods:** In protocol 1, ^{99m}Tc-tetrofosmin, ^{201}Tl and microspheres were injected during total left anterior descending (LAD) coronary artery occlusion in five anesthetized open-chested dogs. Protocol 2 dogs underwent LAD occlusion for either 60 min (n = 7) or 180 min (n = 6) followed by 105 min of reperfusion. ^{99m}Tc-tetrofosmin (10 mCi), ^{201}Tl (1 mCi) and microspheres were injected 90 min after reflow. In both protocols, myocardial ^{99m}Tc-tetrofosmin and ^{201}Tl activities were quantified from regions of interest on ex vivo images and by in vitro well counting. **Results:** In protocol 1, there was a linear relationship between ^{201}Tl ($r = 0.96$) and ^{99m}Tc-tetrofosmin ($r = 0.92$) activities and microsphere flow during the occlusion. In protocol 2, the LAD/left circumflex (LCx) defect count ratios for ^{99m}Tc-tetrofosmin and ^{201}Tl from images of myocardial slices were comparable in dogs undergoing either 1 or 3 h of LAD occlusion and 105 min of reperfusion. Similarly, the LAD/LCx in vitro count ratios were comparable between ^{201}Tl and ^{99m}Tc-tetrofosmin in 1 and 3 h occluded dogs, and significantly lower than the reperfusion flow when these tracers were injected. Uptake of both tracers was depressed to a greater extent in areas of severe ischemic damage. **Conclusion:** These data suggest that administration of ^{99m}Tc-tetrofosmin during coronary occlusion accurately delineates the flow heterogeneity. When given after reperfusion, ^{99m}Tc-tetrofosmin uptake was significantly reduced in reperfused, infarcted areas and was reflective of viability and the degree of myocardial salvage in addition to reperfusion flow. These experimental studies validate the clinical use of ^{99m}Tc-tetrofosmin for assessing persistent coronary artery occlusion, and infarct size and myocardial viability after reperfusion.

Key Words: ^{201}Tl; ^{99m}Tc-tetrofosmin; radionuclide imaging; myocardial infarction

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Technetium-99m-1,2-bis[bis(2-ethoxyethyl)phosphino]-ethane (tetrofosmin) is a lipophilic cationic complex that exhibits excellent myocardial uptake and rapid blood clearance after an intravenous injection (*1*). Additionally, ^{99m}Tc-tetrofosmin shows slow myocardial clearance with no appreciable delayed redistribution (*1*). This ^{99m}Tc-labeled myocardial perfusion agent has high photon flux and energy, which make it ideally suitable for SPECT. Because of its faster clearance from both lung and liver than ^{99m}Tc-sestamibi, some observers have suggested the possibility of earlier imaging after tracer injection to obtain high quality scintigrams (*2*). ^{99m}Tc-tetrofosmin is taken up by myocardial tissue in proportion to blood flow and myocyte viability (*3,4*); however, ^{99m}Tc-tetrofosmin activity underestimates flow at high flows above 2.0 mL/min/g (*3,4*). Like ^{99m}Tc-sestamibi, ^{99m}Tc-tetrofosmin is bound in mitochondria, with uptake being driven by the electropotential gradient according to the Nernst equation (*5,6*). Uptake of ^{99m}Tc-tetrofosmin is inhibited by metabolic inhibitors and excessive calcium, lending evidence to its use as a viability agent.

In a multicenter trial that enrolled 357 patients, acute rest imaging with ^{99m}Tc-tetrofosmin in the emergency department in patients with chest pain consistent with an acute ischemic syndrome, and who had a nondiagnostic electrocardiogram, yielded a 98% negative predictive value of a normal scan for acute infarction (*7*). Patients with an abnormal scan had significantly more acute myocardial infarction (21% versus 2%) and more revascularization (23% versus 7%) than patients with a normal scan. The size of the defect area in patients with positive scans, however, was not assessed regarding its prognostic importance.

Previous experimental and clinical studies have shown that because of minimal redistribution, myocardium at risk can be accurately quantified with resting ^{99m}Tc-sestamibi perfusion imaging when the tracer is injected during acute coronary occlusion (*8–13*). In addition, ^{99m}Tc-sestamibi administered after coronary reperfusion is useful for assessing the degree of myocardial salvage attained with reflow. Infarct size as measured with ^{99m}Tc-sestamibi imaging is predictive of prognosis (*14*).

The purpose of this study was to determine whether ^{99m}Tc-tetrofosmin, which may have a lower extraction fraction than ^{99m}Tc-sestamibi (*4*), can also accurately quantify the regional flow heterogeneity when injected during acute coronary occlusion. If this were true, ^{99m}Tc-tetrofos-

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min imaging might be clinically useful as a triage tool for detecting persistent coronary occlusion. In addition, because ^{99m}Tc-tetrofosmin clears slowly from the myocardium and does not undergo significant redistribution, a myocardial distribution proportional to blood flow over the low-flow range would suggest that ^{99m}Tc-tetrofosmin might also be useful for assessing the myocardial area at risk when the tracer is injected before reperfusion therapy. Finally, in this study we also examined whether ^{99m}Tc-tetrofosmin imaging could be used to estimate the degree of myocardial salvage and viability after coronary reperfusion. If this agent is to be clinically used in the acute care setting in patients with acute infarction, further experimental data validating such applications appears warranted.

MATERIALS AND METHODS

Surgical Preparation

The surgical preparation used for these experiments has been previously described (*9*). Eighteen dogs (mean weight 21.7 ± 1.0 kg) were anesthetized with sodium pentobarbital (30 mg/kg), intubated and ventilated on a respirator (Harvard Apparatus, South Natick, MA). The left femoral vein was cannulated with an 8F polyethylene catheter for administration of fluids, medications and radionuclides. Both femoral arteries were isolated and cannulated with 8F polyethylene catheters for blood pressure monitoring, collection of arterial blood samples and microsphere reference blood withdrawal. The ventilator was adjusted and bicarbonate administered to maintain blood gases within the normal physiological range.

A thoracotomy was performed at the fifth intercostal space, and the heart was suspended in a pericardial cradle. A flare-tipped polyethylene catheter was inserted into the left atrial appendage for pressure measurement and injection of microspheres. Proximal segments (1.5-cm) of both the left anterior descending (LAD) artery and the left circumflex artery (LCx) were dissected free of the epicardium. Ultrasonic flow probes (T201; Transonic Systems Inc., Ithaca, NY) were placed on the LAD and LCx coronary arteries. A snare ligature was then placed proximal to the probe. An eight-channel stripchart recorder (Model 7758A; Hewlett-Packard Co., Lexington, MA) was used to monitor lead II of the electrocardiogram, arterial and left atrial pressures and ultrasonic LAD and LCx flows. All experiments were performed with the approval of the University of Virginia Animal Research Committee, in compliance with the position of the American Heart Association on use of research animals.

Preparation of ^{99m}Tc-Tetrofosmin

^{99m}Tc-tetrofosmin was prepared from a freeze-dried kit provided in a 10-mL glass vial stored at 2°–8°C. The vial was reconstituted with 5 mL of a sodium pertechnetate solution by diluting the eluate from a ^{99m}Tc generator with 0.9% saline. The vial was then shaken gently to ensure complete dissolution of the lyophilized powder and allowed to stand at room temperature for 15 min. Using thin-layer chromatography, mean radiochemical purity was 95.5%.

Experimental Protocols

Protocol 1: Assessment of Flow Heterogeneity During Sustained Occlusion. Protocol 1 (Fig. 1, top) was designed to test whether ^{99m}Tc-tetrofosmin may be used to accurately assess the regional flow heterogeneity when the tracer was injected during sustained total coronary occlusion. After baseline hemodynamic measurements, the LAD arteries of five dogs were totally occluded for 60 min to allow for the recruitment of collateral vessels. ^{99m}Tc-tetrofosmin (5 mCi), ^{201}Tl (1 mCi) and microspheres were administered simultaneously. The microspheres used in these experiments were either ^{113}Sn, ^{103}Ru, ^{95}Nb or ^{46}Sc. Five minutes later, the dogs were killed with an overdose of sodium pentobarbital.

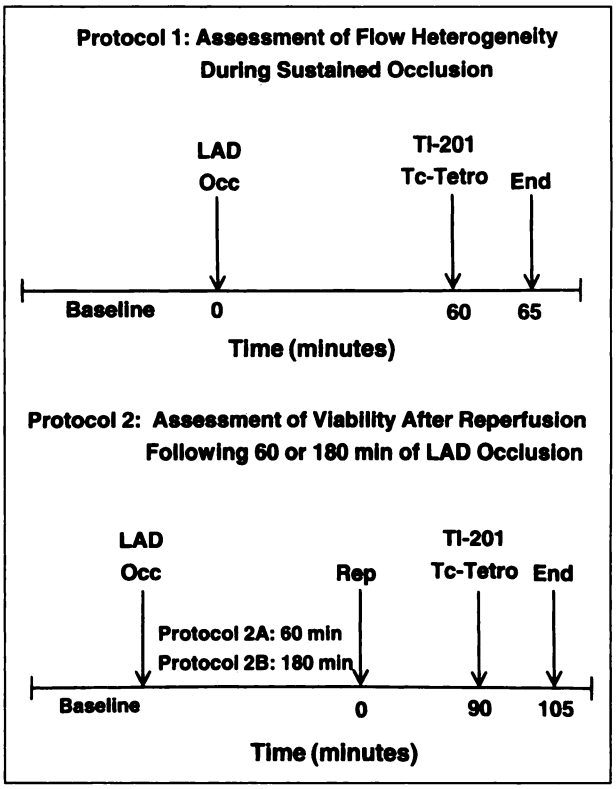

FIGURE 1. Experimental protocols. LAD = left anterior descending coronary artery; Occ = occlusion; Tc-Tetro = ^{99m}Tc-tetrofosmin; Rep = reperfusion.

Protocol 2: Estimation of Myocardial Viability. Protocol 2 was designed to examine the feasibility of using ^{99m}Tc-tetrofosmin for the estimation of myocardial viability using a model of coronary occlusion and reperfusion (Fig. 1, bottom). After baseline hemodynamic measurements, the LAD arteries of 13 dogs were totally occluded for either 60 min (protocol 2A, n = 7) or 180 min (protocol 2B, n = 6) followed by 105 min of reperfusion. Ninety minutes after reperfusion, all 13 dogs were injected with ^{99m}Tc-tetrofosmin (10 mCi) and ^{201}Tl (1 mCi). Radiolabeled microspheres were administered to assess regional myocardial blood flow at baseline, during occlusion and after reperfusion at the time when the tracers were administered. At the end of the protocol, before killing the dogs, the LAD artery was reoccluded, and monastral blue dye was injected into the left atrial catheter to define the anatomic risk area. The dogs then were killed with an overdose of sodium pentobarbital.

Postmortem Analysis

At the end of the experiment, the heart was excised and sliced evenly from apex to base into four segments as previously described (*9*). The left ventricle and septum then were separated from the remainder of the heart and photographed. Each slice was carefully traced on acetate sheets to define the endocardial and epicardial borders and the area at risk. Protocol 2 heart slices were

subsequently incubated in 1% phosphate-buffered triphenyl tetrazolium chloride (TTC) solution for 10 min to define infarct size, then rephotographed and retraced on the same acetate sheets. Using a digital planimeter program (DigiPlan; Scientific Computing Solutions, Charlottesville, VA), risk area and infarct area were determined.

Determination of Regional Myocardial Blood Flow Using Radioactive Microspheres and Quantification of Tracer Uptake

The technique used in our laboratory for quantification of regional myocardial blood flow using radioactive microspheres has been previously described (*4*). A dose of spheres (2–4 million; mean diameter, 11 μm) was suspended in 10% dextran and Tween 80. Normal saline was added to bring the total volume to 3 mL. Uniform mixing was accomplished initially by mechanical agitation (Vortex Genie mixer; Scientific Industries Inc., Bohemia, NY), followed by hand agitation between two syringes attached by a three-way stopcock. The microspheres were administered over 15 s into the left atrium. For flow determination, paired arterial reference samples were obtained by continuous withdrawal (Harvard Apparatus) over 130 s, beginning 10 s before the injection of each set of spheres. Each of the four myocardial slices was divided into eight transmural sections, which were further subdivided into epicardial, midwall and endocardial segments, resulting in a total of 96 myocardial segments for each dog. The myocardial segments and arterial blood samples were counted for ^{99m}Tc-tetrofosmin activities in a gamma-well scintillation counter (MINAXI 5550; Packard Instruments, Downers Grove, IL) within 24 h using a window setting of 120–160 keV. Forty-eight hours later, when the ^{99m}Tc activity had decayed, the myocardial and blood samples were counted for ^{201}Tl activity using a window setting of 50–100 keV. Three weeks later, when ^{201}Tl activity was negligible, the myocardial samples were counted a third time for microsphere regional blood flow determination. The microsphere window settings were: ^{113}Sn, 340–440 keV; ^{103}Ru, 450–550 keV; ^{95}Nb, 640–840 keV; and ^{46}Sc, 842–1300 keV. Tissue counts were corrected for background, decay and isotope spillover, and regional myocardial blood flow was calculated using computer software developed for this purpose (PCGERDA; Scientific Computing Solutions). The transmural regional flow values for a specific sample were derived from the weighted average of epicardial, midwall and endocardial values for that sample. To facilitate comparisons between tracer activity and flow, the ^{201}Tl and ^{99m}Tc-tetrofosmin activities and microsphere flows were normalized to the average value of 15–18 samples taken from the nonischemic region supplied by the LCx.

Image Acquisition and Quantification of Defect Count Ratio

The heart slices were placed on a piece of thin cardboard and covered with plastic wrap. The slices were imaged directly on the collimator of a standard nuclear medicine gamma camera and computer (Technicare 420; Ohio Nuclear, Solon, OH). An all-purpose, low-to-medium energy collimator with a 20% window centered around the photopeak of ^{99m}Tc and a 25% window centered around the photopeak of ^{201}Tl were used. The heart slices were imaged on the day of the experiment for ^{99m}Tc activity. The next day, when the ^{99m}Tc had significantly decayed, the heart slices were reimaged using the ^{201}Tl window settings.

For quantification of ^{99m}Tc activity, regions of interest were drawn on the defect area of the anteroseptal left ventricular wall, and on the normally perfused posterior wall of each image. Regions of interest were drawn to encompass as great an amount of the region in question without including border areas. The ex vivo defect count ratio was computed by dividing the average counts per pixel in the ischemic region by the average counts per pixel in the nonischemic region.

Statistical Analysis

Computations were performed using Systat software (SPSS; Systat Inc., Evanston, IL). Results are expressed as mean values ± 1 SEM. Differences were determined using univariate repeated measures analysis of variance, followed by appropriate post hoc comparisons. Differences were considered significant at a P value of <0.05 (2-tailed). Linear regression analyses were also performed on the tracer activity versus flow data in both protocols using the general linear modeling algorithms within Systat.

RESULTS

Protocol 1

Hemodynamics. The hemodynamics at baseline and during acute coronary occlusion for dogs undergoing acute LAD artery occlusion in protocol 1 are shown in Table 1.

Note that there was a slight but insignificant reduction in heart rate, a stable mean arterial pressure and an expected reduction in LAD arterial flow (from 17 mL/min to 0 mL/min).

Uptake of ^{99m}Tc-Tetrofosmin and ^{201}Tl Versus Microsphere Flow. Figure 2 shows the myocardial uptake of ^{201}Tl and ^{99m}Tc-tetrofosmin versus occlusion flow as determined by microspheres in all protocol 1 dogs.

The tracer activity and flow values from the individual dogs were combined after normalization to the activity at a normal flow of 1.0 mL/min/g. Note that for both ^{201}Tl (r = 0.96) and ^{99m}Tc-tetrofosmin (r = 0.92), there was a linear relationship between tracer activities and microsphere flow. The relative uptake of the two tracers was similar over the low to normal flow range.

TABLE 1
Mean Hemodynamic Parameters

	Group	Baseline	Occlusion	Reperfusion
Protocol 1 dogs				
HR (beats/min)		125 ± 3	115 ± 3	
AP (mm Hg)		94 ± 3	92 ± 6	
LAP (mm Hg)		9 ± 1	9 ± 1	
LAD flow (mL/min)		17 ± 3	0 ± 0*	
Protocol 2 dogs				
HR (beats/min)	2A	115 ± 5	113 ± 5	103 ± 4
AP (mm Hg)	2A	99 ± 5	92 ± 5	90 ± 4
LAP (mm Hg)	2A	10 ± 1	11 ± 1	9 ± 1
LAD flow (mL/min)	2A	18 ± 2	0 ± 0*	18 ± 1
HR (beats/min)	2B	118 ± 7	109 ± 5	110 ± 4
AP (mm Hg)	2B	104 ± 6	98 ± 3	84 ± 4*
LAP (mm Hg)	2B	10 ± 1	15 ± 1*	15 ± 1*
LAD flow (mL/min)	2B	24 ± 5	0 ± 0*	20 ± 3

*$P < 0.01$ versus respective baseline. All values are mean ± SEM.
Group 2A = 60 min occlusion; Group 2B = 180 min occlusion.
HR = heart rate; AP = arterial pressure; LAP = left atrial pressure; LAD = left anterior descending artery.

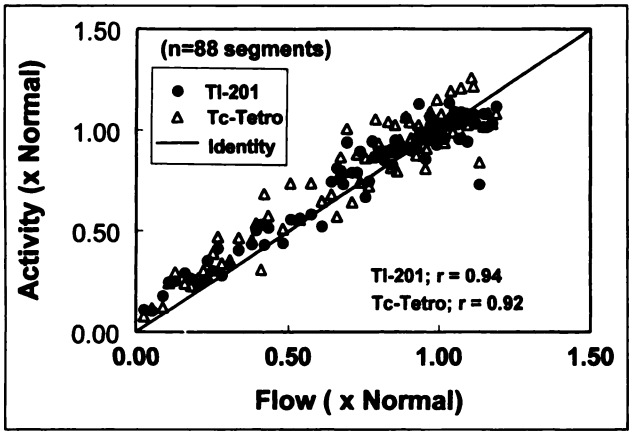

FIGURE 2. Plots of normalized ^{201}Tl and ^{99m}Tc-tetrofosmin (Tc-Tetro) activities versus flow during LAD coronary artery occlusion. Activities and flow values were normalized to average activity and flow in normal segments from left circumflex region. For purposes of clarity, only every fifth point is displayed.

Ex Vivo Imaging of Heart Slices. As mentioned previously, regions of interest were drawn in the defect area in the anteroseptal region of the left ventricular wall, for quantification of ex vivo defect count ratio (defect/normal).

Figure 3 is an example of ex vivo images of the heart slices with ^{201}Tl and ^{99m}Tc-tetrofosmin in a representative dog in protocol 1. Note that the ^{201}Tl and ^{99m}Tc-tetrofosmin defect areas are virtually identical. The ex vivo defect count ratio for all five dogs in this group was 0.32 ± 0.07 for ^{99m}Tc-tetrofosmin and 0.36 ± 0.06 for ^{201}Tl ($P < 0.05$). The mean anatomic risk area by monastral blue dye staining was 23%.

Protocol 2

Hemodynamics. Hemodynamic data for dogs in protocols 2A and 2B that underwent either 60 min or 180 min of coronary occlusion followed by reperfusion, respectively, are shown in Table 1.

After coronary reperfusion in both groups of dogs, mean heart rate was unchanged, as was the left atrial pressure. In protocol 2B, dogs that underwent 3 h occlusion before reflow, mean arterial pressure fell from 98 ± 3 to 84 ± 4 mm Hg ($P < 0.01$). Also in protocol 2B dogs, the left atrial pressure rose significantly after coronary occlusion, from 10 ± 1 to 15 ± 1 mm Hg and remained elevated after 105 min of coronary reperfusion at 15 ± 1 mm Hg. For both groups, LAD flow returned to baseline after reperfusion. During occlusion, flow fell to 0 mL/min.

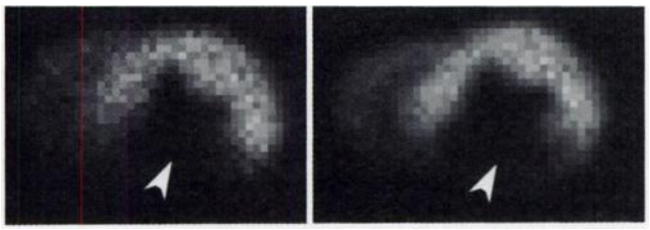

FIGURE 3. Representative ^{201}Tl and ^{99m}Tc-tetrofosmin ex vivo images of heart slices from a protocol 1 dog. Note that size of myocardial perfusion defect (white arrow) was comparable between ^{201}Tl and ^{99m}Tc-tetrofosmin.

Regional Myocardial Blood Flow. Microsphere blood flow in the endocardial, midwall, epicardial and transmural regions for both the reperfused LAD and normal LCx zones is shown in Table 2.

In both protocols 2A and 2B, regional myocardial blood flow was constant in the normal, LCx zone during baseline, occlusion and after reperfusion. In the LAD zone, there was a similar degree of flow reduction in both protocols 2A and 2B, with endocardial flow values of 0.10 ± 0.01 mL/min/g in both groups. Ninety minutes after reperfusion, at the time when ^{99m}Tc-tetrofosmin and ^{201}Tl were injected, transmural LAD zone flow was elevated relative to baseline and LCx zone flow in both protocols 2A and 2B, indicating persistent hyperemia in the reperfused zone.

Infarct Size. Postmortem infarct size as determined by triphenyl tetrazolium chloride staining is shown in Figure 4 for the protocol 2A dogs undergoing 60 min of occlusion and 105 min of reflow and for protocol 2B dogs undergoing 180 min of occlusion followed by 105 min of reflow.

The anatomic risk area was similar for the protocol 2A (28% of the left ventricle) and protocol 2B (30% of the left ventricle) dogs. Mean infarct size was significantly larger ($P = 0.018$) for dogs with the longer period of occlusion before reflow. Infarct size averaged 3% ± 1% of the left ventricle (12% of risk area) in the protocol 2A dogs, compared with 12% ± 3% of the left ventricle (29% risk area) in the protocol 2B dogs with prolonged occlusion followed by reperfusion.

Myocardial Uptake of ^{99m}Tc-Tetrofosmin and ^{201}Tl After Reperfusion. Figure 5A plots ^{99m}Tc-tetrofosmin activity versus microsphere flow at the time after reperfusion when the tracer was injected in the protocol 2B dogs.

The tissue activity values from each dog in this group were normalized to the activity at a normal flow of 1.0 mL/min/g. The tissue samples that were TTC positive (viable) are shown as crosses; the TTC-negative (nonviable) samples are shown as triangles. Note that there was a poor correlation ($r = 0.33$) between ^{99m}Tc-tetrofosmin activity and reperfusion flow, resulting mainly from the diminished uptake of ^{99m}Tc-tetrofosmin in the nonviable samples. Note also that the diminished ^{99m}Tc-tetrofosmin uptake did not result from inadequate perfusion, because in most of these nonviable segments flow was normal or even hyperemic. In Figure 5B, the same tissue samples are plotted as a function of flow during the LAD occlusion. Note that there was a high correlation between ^{99m}Tc-tetrofosmin activity and occlusion flow ($r = 0.86$), despite the fact that the tracer was injected after reperfusion, indicating that the myocardial uptake of ^{99m}Tc-tetrofosmin is dependant on myocyte viability as well as flow.

Figure 6A shows the LAD-to-LCx artery ratio of ^{201}Tl uptake, ^{99m}Tc-tetrofosmin uptake and reperfusion flow in

TABLE 2
Regional Myocardial Blood Flow

	LAD zone (reperfused)			LCx zone (normal)		
	Baseline	Occlusion	Reperfusion	Baseline	Occlusion	Reperfusion
Protocol 2A						
Endo	0.79 ± 0.03	0.10 ± 0.01*	1.58 ± 0.18*	0.96 ± 0.01	0.96 ± 0.01	0.99 ± 0.01
Mid	0.85 ± 0.03	0.18 ± 0.02*	0.97 ± 0.06	0.97 ± 0.01	0.94 ± 0.01	0.99 ± 0.01
Epi	1.16 ± 0.06	0.42 ± 0.04*	1.38 ± 0.07†	1.06 ± 0.02	1.06 ± 0.03	1.08 ± 0.03
TM	0.94 ± 0.03	0.23 ± 0.03*	1.28 ± 0.07*	0.99 ± 0.02	0.99 ± 0.02	1.02 ± 0.01
Protocol 2B						
Endo	0.80 ± 0.04	0.10 ± 0.02*	1.50 ± 0.27	0.98 ± 0.01	0.95 ± 0.02	0.99 ± 0.02
Mid	0.84 ± 0.04	0.16 ± 0.05*	1.03 ± 0.11	0.98 ± 0.01	0.94 ± 0.02	0.98 ± 0.02
Epi	1.15 ± 0.10	0.35 ± 0.09*	1.29 ± 0.09	1.06 ± 0.03	1.03 ± 0.06	1.05 ± 0.04
TM	0.93 ± 0.05	0.20 ± 0.06*	1.24 ± 0.11†	1.01 ± 0.01	0.97 ± 0.03	1.00 ± 0.02

*$P \leq 0.01$.
†$P < 0.05$ vs. baseline.
Protocol 2A = 60 min LAD occlusion; Protocol 2B = 180 min LAD occlusion.
LAD, LCx = left anterior descending, left circumflex coronary arteries; endo = endocardial samples; mid = midwall samples; epi = epicardial samples; TM = transmural samples.

transmural myocardial segments obtained from protocol 2A dogs that underwent 60 min of LAD artery occlusion followed by 105 min of reflow. Myocardial segments were grouped according to the severity of the flow reduction during the LAD artery occlusion. Normal segments had 60% or more of normal LCx arterial flow, moderate segments had 30% or more and less than 60% and severe segments had less than 30% normal flow. Note that in normal segments, in segments with a moderate flow reduction and in segments with a severe flow reduction, ^{201}Tl uptake and ^{99m}Tc-tetrofosmin uptake were comparable and significantly less than reperfusion flow. Similarly, in protocol 2B dogs that were occluded for 3 h before reperfusion (Fig. 6B), the LAD-to-LCx count ratios were comparable between ^{201}Tl and ^{99m}Tc-tetrofosmin and significantly lower than the reperfusion flow when these tracers were injected. Uptake of both tracers was depressed to a greater extent in areas of severe ischemia. Thus, in both groups of dogs, ^{201}Tl and ^{99m}Tc-tetrofosmin uptake during reperfusion were more reflective of viability and the degree of myocardial salvage than reflective of reperfusion flow.

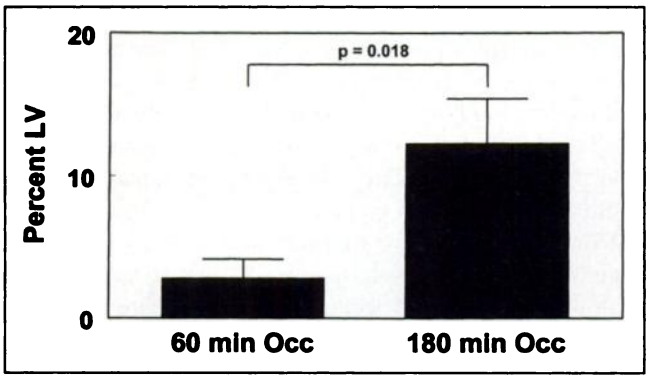

FIGURE 4. Comparison between myocardial infarct sizes (percent left ventricle [LV]) in protocol 2 dogs that underwent either 60 min (protocol 2A) or 180 min (protocol 2B) of total LAD artery occlusion (Occ) followed by reperfusion. Myocardial risk area was comparable between the two groups, but infarct size was significantly higher in protocol 2B dogs.

Image Defect Count Ratios. The LAD-to-LCx defect count ratios from images of the myocardial slices for ^{201}Tl and ^{99m}Tc-tetrofosmin uptake are shown in Figure 7.

As shown, uptake of the two tracers was comparable in both protocol 2A and 2B dogs, with a significantly lower uptake in the latter group, which had 3 h of occlusion before reflow. These data are consistent with the in vitro well counting data shown in Figure 6.

DISCUSSION

The findings of this study suggest that after the administration of ^{99m}Tc-tetrofosmin during coronary artery occlusion in dogs, the relative distribution of the radiolabeled tracer accurately delineates the perfusion heterogeneity. When ^{99m}Tc-tetrofosmin was given after 90 min of reperfusion, after either 1 or 3 h of coronary occlusion, the myocardial uptake of the tracer was significantly reduced in infarcted areas, compared with reperfusion flow, hence reflecting the degree of myocardial salvage and viability rather than just the degree of flow restoration. Furthermore, ^{99m}Tc-tetrofosmin uptake was comparable to ^{201}Tl uptake in all groups of dogs. Our finding that ^{99m}Tc-tetrofosmin uptake accurately delineates the occlusion flow heterogeneity suggests that this tracer would be well suited for the noninvasive assessment of vessel patency; i.e., as a triage tool in the setting of persistent coronary occlusion. Importantly, as has been previously observed with ^{99m}Tc-sestamibi (*8–14*), the findings in this study support the concept that serial ^{99m}Tc-tetrofosmin imaging during acute myocardial infarction can delineate the area of myocardium at risk during occlusion

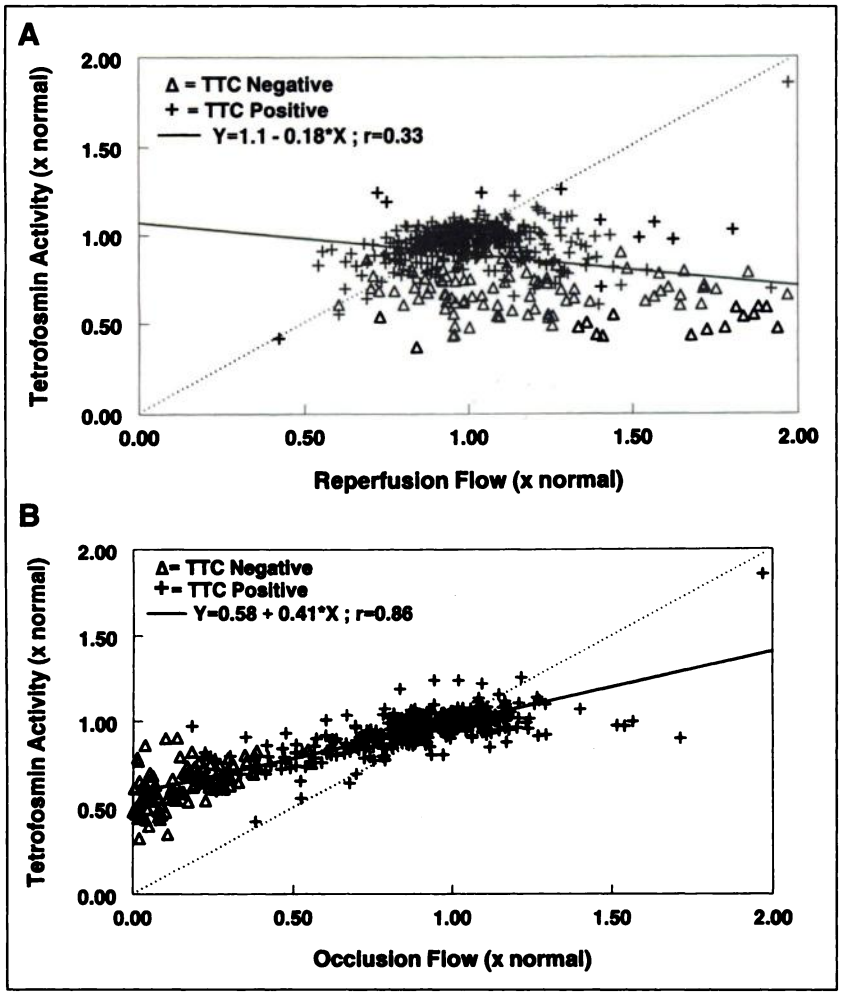

FIGURE 5. (A) ^{99m}Tc-tetrofosmin activity plotted as function of reperfusion flow from protocol 2B dogs. Although ^{99m}Tc-tetrofosmin was injected after reperfusion, there was poor correlation (r = 0.33) between tracer activity and reperfusion flow because of reduced uptake of tracer in nonviable segments despite normal or hyperemic flow at time of injection. Dotted line represents line of identity. (B) ^{99m}Tc-tetrofosmin activity plotted as function of flow during LAD occlusion from same dogs as in panel A. ^{99m}Tc-tetrofosmin activity was highly correlated (r = 0.86) with occlusion flow, despite tracer being injected after reperfusion, indicating that tracer uptake is dependent on myocardial viability in addition to flow.

and subsequently provide an assessment of degree of salvage and infarct size after reflow.

Previous Studies

The findings of this study are consistent with other data previously reported in the literature regarding ^{99m}Tc-tetrofosmin. Takahashi et al. (*15*) showed that in rats undergoing 1 h of occlusion and 1 h of reperfusion of a major branch of the LCx artery, the initial uptake and delayed (1 h) retention of ^{99m}Tc-tetrofosmin were affected by the status of myocardial viability. There were significantly lower uptake and retention values for ^{99m}Tc-tetrofosmin when nonviable segments were compared with viable segments at all flow levels. These data are also consistent with our previous observations showing substantial ^{99m}Tc-tetrofosmin uptake (>50% of nonischemic) in dogs with a chronic reduction in flow causing ischemic dysfunction but sustained viability (*16*). ^{99m}Tc-tetrofosmin accumulates in mitochondria similar to ^{99m}Tc-sestamibi, and, as shown in this study, only viable myocardial tissue sequesters the tracer. Its myocardial uptake is reduced by metabolic inhibitors (such as iodoacetic acid and 2,4-dinitrophenol) and excess of influx of calcium (*5,6*).

Assessment of Coronary Reperfusion

A promising alternative to ^{201}Tl scintigraphy for assessment of reperfusion therapy in patients is the administration of ^{99m}Tc-sestamibi before and after reflow. Because this agent does not redistribute, the first dose can be administered just before thrombolytic therapy, but imaging can be postponed several hours later after thrombolytic drug administration is complete to obtain the pretreatment assessment of risk area. A "snapshot" of the risk area at the time of admission is obtained with this first injection. A second injection is then administered sometime after reperfusion and after the first pretreatment images are obtained, to delineate the extent of myocardial salvage (*10,11*). In clinical studies, patients with a patent infarct artery had a significantly greater decrease in defect size on repeat images performed 18–48 h after thrombolytic therapy, than did patients with persistently occluded vessels (*10,11*). Wackers et al. (*10*) found that a relative decrease of > 30% in the size of the ^{99m}Tc-sestamibi perfusion defect predicted patency of the infarct-related artery, providing noninvasive evidence of the efficacy of treatment. The results of early ^{99m}Tc-sestamibi imaging in postinfarction patients receiving throm-

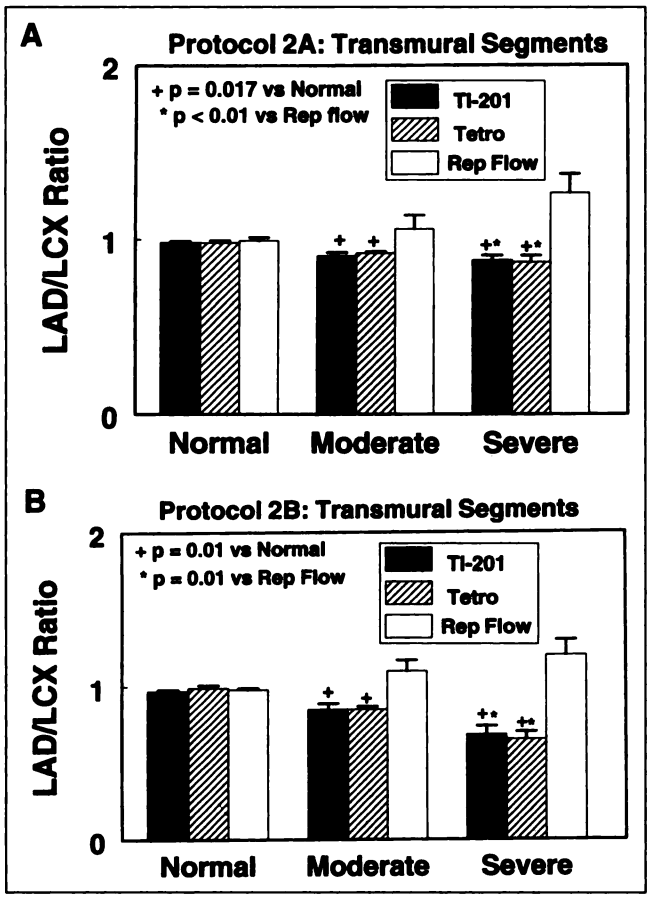

FIGURE 6. (A) Transmural LAD/LCx artery ^{201}Tl (solid bars), ^{99m}Tc-tetrofosmin (Tetro) activity (striped bars) and reperfusion (Rep) flow (open bars) ratios in Protocol 2A dogs (60 min occlusion and Rep). ^{201}Tl and tetro activities were comparable and were significantly lower in moderate to severe ischemic regions than in normal regions. Both tracers underestimated Rep flow in most severe segments. (B) Transmural activity and Rep flow ratios from protocol 2B dogs (180 min occlusion and Rep). Further reduction in tracer activity ratios in moderate-severe regions reflects greater extent of myocardial injury present in this group. Again note that ^{201}Tl and tetro activities were comparable in all three regions.

bolytic therapy are also predictive of late functional improvement (*17*). ^{99m}Tc-sestamibi or ^{99m}Tc-tetrofosmin imaging could become a useful technique to incorporate in clinical research trials aimed at evaluating efficacy of interventions directed at restoring blood flow after acute coronary occlusion (e.g., primary angioplasty) (*18–20*). Behrenbeck et al. (*18*) found a significant reduction in defect size after primary angioplasty in patients with a first transmural infarction who underwent serial ^{99m}Tc-sestamibi imaging. Gibbons et al. (*19*) found that myocardial salvage as assessed by measuring the percent reduction in ^{99m}Tc-sestamibi defect size was similar in patients randomized to primary angioplasty and patients randomized to thrombolytic therapy. Clements et al. (*20*) found that residual antegrade flow before direct angioplasty and demonstration of collateral flow were associated with a significantly smaller, final ^{99m}Tc-sestamibi SPECT defect size and more myocardial salvage after angioplasty.

In this study, ^{99m}Tc-tetrofosmin activity accurately delineated the occlusion flow heterogeneity. Thus, ^{99m}Tc-tetrofosmin imaging may be well suited for use as a triage tool for assessing persistent coronary occlusion after unsuccessful reperfusion therapy. In addition, because of insignificant redistribution of ^{99m}Tc-tetrofosmin postinjection, there is no apparent reason why this tracer cannot achieve the same results as described with ^{99m}Tc-sestamibi in the assessment of myocardium at risk. Similarly, ^{99m}Tc-tetrofosmin after reperfusion was reflective of the amount of myocardium salvaged, which is similar to what was reported with ^{99m}Tc-sestamibi previously (*9*). However, in this study, ^{99m}Tc-tetrofosmin and ^{201}Tl were injected at 90 min after reperfusion rather than at 180 min as in the previous study (*9*). Thus, the higher activity ratios in the severe regions likely reflect a greater amount of postreperfusion hyperemia in the current studies. Nevertheless, ^{99m}Tc-tetrofosmin uptake was also comparable to ^{201}Tl uptake after reflow, further supporting the conclusion that viability is being assessed with this agent. ^{201}Tl imaging has been validated for determination of myocardial viability (*21–23*).

Thus, the major clinical implication of this experimental study is that ^{99m}Tc-tetrofosmin can be substituted for ^{201}Tl for the assessment of regional flow heterogeneity during coronary occlusion, and infarct size and residual myocardial viability in patients with acute myocardial infarction. The data indicate that the uptake of ^{99m}Tc-tetrofosmin was comparable to the uptake of ^{201}Tl, both in dogs undergoing 1 h of occlusion and in dogs undergoing 3 h of occlusion before reflow, and the uptake of both tracers was significantly less than reperfusion flow. Defect size, like ^{99m}Tc-sestamibi defect size, should have significant prognostic import in the clinical setting. The larger the defect size, reflective of infarct size, the worse the long-term outcome and the higher the mortality rate. Further clinical trials with

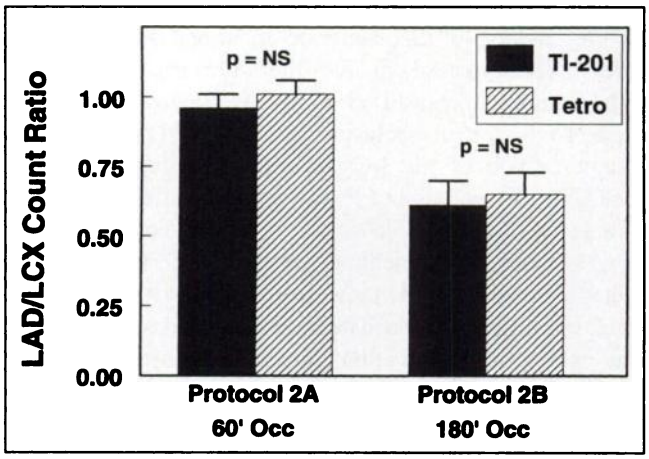

FIGURE 7. ^{201}Tl (solid bars) and ^{99m}Tc-tetrofosmin (Tetro, striped bars) LAD/LCx artery mean defect count ratios from ex vivo image in protocol 2A (60 min LAD occlusion [Occ] and reperfusion) and protocol 2B (180 min Occ and reperfusion) dogs. By quantitative imaging, ^{201}Tl and Tetro were comparable for detecting myocardial infarction in both groups of dogs.

^{99m}Tc-tetrofosmin imaging in patients with acute myocardial infarction appear warranted to further validate its use in the clinical setting for assessment of risk area and viability.

STUDY LIMITATIONS

One limitation of this study is that the dogs in protocol 1 were not reperfused. Therefore, it is unknown whether the occlusion flow heterogeneity would have remained fixed after reperfusion and therefore allow delayed assessment of the myocardial risk area. However, based on the current knowledge of ^{99m}Tc-tetrofosmin kinetics, i.e., slow myocardial clearance with little or no redistribution, it is expected that the postreperfusion ^{99m}Tc-tetrofosmin distribution would be an accurate marker of the myocardial risk area, similar to what has been previously observed with ^{99m}Tc-sestamibi. A second limitation was the degree of hyperemia present at the time when ^{99m}Tc-tetrofosmin and ^{201}Tl were injected at 90 min postreperfusion. The higher than normal flow in the reperfused area at the time of injection may have resulted in an overestimation of the degree of myocardial salvage. Nevertheless, even with the hyperemia present in these studies, the myocardial uptake of ^{99m}Tc-tetrofosmin was substantially lower than reperfusion flow and was not significantly different from that of ^{201}Tl. It should also be emphasized that hyperemia at the time of reperfusion affects the myocardial uptake of all perfusion tracers and may result in the overestimation of the amount of myocardium salvaged. Thus, this factor should be considered when designing clinical protocols to assess myocardial viability early after reperfusion therapy.

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