Nitrogen-13-Ammonia and PET to Detect Allograft Coronary Artery Disease after Heart Transplantation: Comparison with Coronary Angiography

Xiao-Ming Zhao, Dominique Delbeke, Martin P. Sandler, Tiong-Keat Yeoh, John R. Votaw and William H. Frist

Vanderbilt Transplant Center and Departments of Cardiothoracic Surgery, Radiology and Radiological Sciences and Medicine, Vanderbilt University Medical Center, Nashville, Tennessee

The diffuse nature of allograft coronary artery disease (CAD) suggests that global myocardial blood flow (MBF) may decrease with time after transplantation; therefore the diagnosis of this disease remains problematic. **Methods:** To investigate whether PET detects a fall in allograft MBF over time, PET scans (108) were obtained from 43 heart transplant recipients. Thirty-five patients underwent two serial PET scans 1 yr apart. MBF was measured by PET using ^{13}N-ammonia as a tracer. Coronary angiography was performed parallel with PET imaging and compared with perfusion rates measured by PET scans. **Results:** MBF measured by PET decreased sequentially with time. The mean MBF was 73 ± 21, 56 ± 13, 51 ± 11 and 51 ± 27 ml/min/100 g of tissue in patients surviving 3 mo, 1, 2 and 3 yr after transplantation, respectively. Significant MBF decrease occurred within 1 yr after transplantation. Sequential PET studies showed a decrease in MBF in 22 of 35 patients (63%). Mean MBF for the first and second scans was 65 ± 18 and 54 ± 16, respectively. MBF decrease was more profound in patients (n = 11) angiographic evidence of CAD. There was a trend towards increased rejection and CMV infection rates in patients with decreased MBF. **Conclusion:** With time, PET detects a decrease in MBF in cardiac allografts. The frequency of MBF decrease detected by PET is concordant with the true incidence of allograft CAD, suggesting that sequential PET is a more sensitive modality for monitoring allograft CAD than angiography.

Key Words: positron emission tomography; cardiac transplantation; coronary artery disease; nitrogen-13-ammonia; coronary angiography

J Nucl Med 1995; 36:982–987

Allograft coronary artery disease (CAD) poses a major impediment for long-term heart transplantation. It develops in about 15% of cardiac transplant recipients each year (*1,2*) and is the major cause of later death in heart transplant recipients. Allograft CAD is characterized by diffuse vascular intimal hyperplasia involving the entire length of the epicardial and intramyocardial coronary arteries (*3*). Because of the nature of this disease, its recognition by coronary angiography is limited (*4,5*). Recent studies reported that virtually all cardiac allografts developed vascular lesions 1 yr after transplantation (*6*). In contrast, less than 40% of patients who survived more than 4 yr had angiographic evidence of CAD (*3,4*). Several studies have demonstrated that a considerable number of allografts which had significant CAD (lumen narrowing >80%) determined at autopsy or retransplantation showed no sign of the disease by conventional angiography (*6*). More reliable modalities to detect the development of allograft CAD are clearly desirable. Computerized quantitative coronary angiography (*7*) and intracoronary ultrasound (*8–9*) have been proposed to detect allograft CAD. PET is a noninvasive modality which has been used to assess primary CAD (*10–11*) and cardiac allograft rejection (*12–13*). Flow and metabolic studies of the heart have been accurately quantitated with positron-emitting radiotracers such as ^{13}N-ammonia (*11*) and [^{18}F]fluorodeoxyglucose (*14–15*). The purpose of this study was to evaluate the ability of PET using ^{13}N-ammonia to accurately assess the progression of allograft CAD.

MATERIALS AND METHODS

Patients

Forty-three heart transplant recipients (randomized) were studied by PET and coronary angiography. Thirty-seven men and six women had a mean age of 45 ± 13 yr and the mean follow-up was 41 ± 15 mo. The original reason for cardiac transplantation among these 43 patients was CAD (n = 20), cardiomyopathy (n = 18), congenital heart diseases (n = 4) and valvular heart diseases (n = 1). One hundred and eight PET scans were obtained from 43 heart transplant recipients at yearly intervals after transplantation. Of the 43 patients, 35 underwent sequential studies 1 yr apart. Coronary angiography was performed within 72 hr following all PET

Received Feb. 3, 1994; revision accepted Sept. 30, 1994.
For correspondence or reprints contact: Dominique Delbeke, MD, PhD, Department of Radiology and Radiological Sciences, Vanderbilt University Medical Center, 21st & Garland, Nashville, TN 37232-2675.

studies and was analyzed by staff cardiologists who were unaware of the PET results.

All patients in the study were clinically stable at the time of their PET scan. The ventriculogram showed no evidence of regional wall motion abnormalities and a myocardial biopsy specimen obtained during the catheterization procedure showed no evidence of acute rejection. All patients received standard immunosuppression (cyclosporine, azathioprine and prednisone).

PET Imaging

PET was performed with a Siemens ECAT 933/08/16 tomograph (Siemens Medical System, Inc., Hoffman Estates, IL). The sensitivity, as measured by scanning a 20-cm diameter cylindrical uniform phantom, was 13,000 cps/mCi/ml in the direct planes (1.3.5. . .15) and 23,000 cps/mCi/ml in the cross planes. The axial field of view (FOV) was 12.8 cm, intrinsic resolution was 4.8 mm at the center of the FOV and reconstructed resolution was 7.5 mm (FWHM). An average of 2.5 million true counts per image existed. The scanner hardware estimates and corrects for random events using the standard delayed window technique. Electrocardiographic gating was not used. Transmission images were obtained to correct for photon attenuation. Dynamic image collection was started immediately after intravenous administration of 20 mCi ^{13}N-ammonia. The dynamic sequence typically consisted of a 0–4 min (frame 1) image for blood-pool information, followed by a 4–19 min (frame 2) image for measuring tissue activity distribution.

Quantitation of Regional Myocardial Perfusion

All the PET scans were analyzed by two independent observers, who were blinded to the clinical status of the patient and to the coronary angiogram results. A quantitative index of MBF was calculated and parametric images were generated using the following formula (*16*):

$$\text{Net extraction} = E \times MBF = Cm(t) \Big/ \int_0^{4\ \text{min}} Ca(s)\, ds, \qquad \text{Eq. 1}$$

where E is the extraction fraction of ^{13}N-ammonia by the myocardium and was assumed to be one. MBF is in ml/min/g, Cm(t) is the myocardial tissue ^{13}N-ammonia radioactivity at time t (t = 15 min) in nCi/g (1 ml of myocardium is approximately equal to 1.05 g) and Ca(s) is the blood-pool ^{13}N-ammonia radioactivity as a function of time in nCi/ml; s is the integration variable. The arterial input function Ca(t) was estimated from the 0–4 min (frame 1) using a small circular region of interest in the center of the left ventricular cavity. A factor was used to correct for overestimation of the arterial input function in the 0–4-min image, mainly due to contamination by ^{13}N-metabolites and spillover from myocardial walls. This factor was experimentally determined in our laboratory from data obtained in canine experiments (*17*) where absolute MBF was calculated using different techniques and correlated with microspheres data. Although the increase of the ^{13}N-metabolites is slower in human blood than in canine blood, the difference has been shown to be insignificant (*18*). Therefore, the correction factor obtained in canine experiments was used for humans.

Image Analysis

The image volume was reoriented along the short-axis of the heart in 1-cm thick slices. Each set of short-axis images was analyzed with a semi-automatic edge detection program using constant wall thickness of two pixels on each side of the maximum profile count. Eight equal wedge-shaped regions were divided using the junction of the anterior right ventricular wall with the anterior interventricular septum as the angle of origin. Although individual coronary artery anatomy and blood flow distribution varies, the regional MBF was calculated in four arbitrarily defined segments (septum, anterior, lateral and inferior walls) by averaging values in the appropriate sectors, and expressed in ml/min/100 g of tissue. A global MBF index was calculated by averaging values in all sectors. Values of each patient were corrected by the mean rate-pressure product for the patients in the group and divided by the rate-pressure product for the individual patients (*19*). The decrease in flow between first and second studies was expressed as flow decrease index (FDI):

$$FDI = [MBF(1st - 2nd)/MBF\ (1st + 2nd)/2] \times 100\%. \qquad \text{Eq. 2}$$

Statistical Analysis

Changes in MBF over time in serial PET studies were compared in two groups. Group I included patients with angiographic evidence of CAD (n = 11) and Group II included patients without angiographic evidence of CAD (n = 24). Differences in MBF were also compared in groups of patients with different survival times. Statistical significance between each group was determined using the Student t-test for paired data. To assess the reproducibility of the PET scan analyses, each set of images was analyzed by two independent observers. The two sets of flow decrease indices were compared by the Student t-test for paired data and showed no significant difference (p = 0.76).

RESULTS

Coronary Angiography

Coronary angiography was performed parallel with PET. Angiographic findings were determined by observers who were blinded to the PET results. Although the diagnosis of allograft CAD is limited by angiography (*3–5*), there is no other modality allowing accurate detection of this disease process. Therefore, the patients were divided in two groups according to their angiographic findings. Allograft CAD was diagnosed if there were apparent vessel irregularity and diffuse concentric narrowing on coronary angiography (type B and C lesions) using the Stanford criteria (*4*). These types of lesions have been reported to be more frequent in allograft CAD than primary CAD (*4*). None of the patients had a discrete stenosis (type A lesion) occluding more than 50% of the lumen of a major vessel. Of the 43 patients, 11 (25%) had angiographic evidence of CAD on at least one study, and the remaining 32 had no evidence of CAD on coronary angiography. The two groups were similar in sex, donor, recipient age and mean graft ischemia time (Table 1).

PET

Of the 108 PET scans from 43 patients analyzed in this study, 12 were performed 3 mo after transplantation, 23 at 1 yr, 29 at 2 yr, 31 at 3 yr and 13 over 4 yr. MBF measured by PET decreased sequentially with time. The mean MBF of patients at 3 mo and 1, 2 and 3 yr survival groups was 73 ± 21, 56 ± 13, 51 ± 11 and 51 ± 27 ml/min/100 g tissue, respectively. MBF was significantly lower at 1 yr compared with that in patients immediately after transplanta-

TABLE 1
Patient Demographics

	Group I CAD*	Group II Non-CAD
Sex (M/F)	10/1	21/3
Donor age (yr)	26 ± 9	24 ± 10
Recipient age (yr)	44 ± 13	41 ± 17
Mean follow-up (mo)	42 ± 12	39 ± 15
Ischemia time (min)	141 ± 44	123 ± 60

*Angiographic evidence of CAD.

tion ($p < 0.05$). No significant differences in regional MBF were noted between the different segments of left ventricle (anterior, septum, lateral and inferior walls). All angiograms and PET scans were carefully reviewed and angiographic findings were compared to PET images. The only resting defect was seen in the patient shown in Figure 1.

Sequential PET scans were obtained from 35 patients 1 yr apart. A decrease in MBF was documented in 22 patients (63%). The other patients did not have more than a 10% change from the MBF measured on the first PET scan. The mean MBF of all patients was 65 ± 18 ml/min/100 g tissue for the first scan and 54 ± 16 ml/min/100 g tissue for the second scan a year later ($p = 0.001$) (Table 2). Coronary blood flow decrease was more profound in patients with angiographic evidence of CAD compared to those without (Table 2, Fig. 2). The mean MBF of the first PET scan was similar in patients both with and without angiographic evidence of CAD (67 ± 18 vs. 63 ± 13, p = ns). The mean MBF of the second scan, however, was significantly lower in patients with angiographic evidence of CAD (51 ± 11 versus 56 ± 14, $p < 0.01$). A fall in MBF was found in 8 of 11 (72%) patients with and 14 of 24 (58%) patients without evidence of CAD on coronary angiography.

The mean decrease in MBF between the sequential scans in 35 patients was 10 ± 9 ml/min/100 g tissue (Table 2). The FDI was 9% ± 5%. The decrease in MBF and FDI was more severe in the patients with than in those without angiographic evidence of CAD. The mean MBF decrease was 16 ± 5 versus 7 ± 1 ml/min/100 g of tissue ($p = 0.02$) and FDI was 14 ± 4 versus 5 ± 1%, respectively ($p = 0.02$) (Table 2, Fig. 3).

Correlation of Rejection and Other Risk Factors with CAD

Mechanisms underlying CAD are not completely understood. However, immunological injury is implied in the pathogenesis of CAD. We found previously that rejection episodes induced expression of a potent vascular smooth muscle cell growth factor, acidic fibroblast growth factor (aFGF) and its receptor in cardiac allografts (*20–23*). This finding provides evidence to support the hypothesis that rejection may result in vascular intimal hyperplasia, a characteristic feature of CAD, through regulation of vascular growth factor expression. Therefore, we compared the number of rejection episodes and other potential risk factors in patients with and without significant MBF decrease defined by sequential PET studies. As shown in Table 3, there was a trend towards a higher rate of rejection episodes in patients with MBF decrease (n = 22) compared to those without flow decrease (n = 13), 25% of patients with MBF decrease versus 46% of patients without flow decrease were free from rejection ($p = 0.06$) at 12 mo and 14% versus 27% at 24 mo ($p = 0.08$). A trend towards a higher CMV infection rate was also noted in patients with significant MBF decrease; 35% versus 45% of patients without flow decrease were free from CMV infection at 12 mo ($p = 0.07$). Of the other factors examined, sex, recipient age (44 ± 13 versus 41 ± 17 yr), donor age (26 ± 9 versus 24 ± 10 yr), ischemic time during surgery (141 ± 44 versus 123 ± 60 min), doses of immunosuppressive agents and blood pressure were similar in the two groups (Table 3).

DISCUSSION

Allograft CAD is a major cause of morbidity and mortality in long-term survivors of heart transplantation (*1,2*). Recently, Russell et al. reported that as high as 80% of allograft hearts autopsied and explanted showed evidence of CAD disease (*6*). Nevertheless, detection of allograft CAD remains problematic. Coronary angiography, the recognized gold standard for diagnosis of primary CAD, has been shown to be insensitive for detecting allograft CAD because of smooth and diffuse nature (*3–5*). Despite this limitation, patients in this study were divided into two groups according to angiographic evidence of CAD. As previously reported (*3–5*), we found that the angiographic lesions seen in the heart transplant patients with angiographic evidence of CAD were vessel irregularity and diffuse concentric narrowing (type B and C lesions), rather than discrete stenoses of major vessels (type A lesion). In our experience, progression of diffuse intimal thickening may worsen or mask vessel irregularity. Therefore, sequential angiograms may actually show progression of

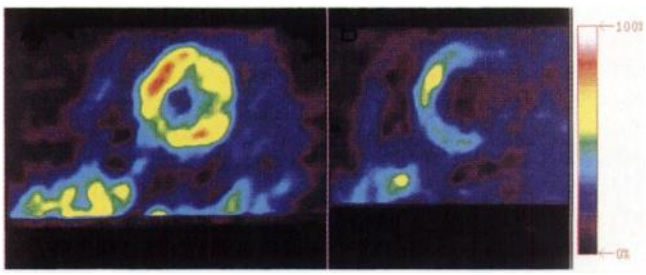

FIGURE 1. A 52-yr-old man 6 yr status postcardiac transplantation. (A) Nitrogen-13-ammonia initial PET scan 5 yr post-transplantation and a short-axis view of the heart show decreased perfusion in the lateral wall. Cardiac catheterization from the same week shows virtually normal coronary arteries. (B) Nitrogen-13-ammonia second PET scan 1 yr later and a short-axis view of the heart show global decreased perfusion and virtually no perfusion to the lateral wall. Cardiac catheterization from the same week shows 90% occlusion of the left circumflex vessel.

TABLE 2
Myocardial Blood Flow Changes with Time in Sequential PET Scans

Category	No.	MBF 1st scan (ml/min/100 g)	MBF 2nd scan (ml/min/100 g)	MBF Decrease (ml/min/100 g)	FDI (%)
Total patients	35	65 ± 18*	54 ± 16*	10 ± 9	9 ± 5
Patients with angiographic CAD	11	67 ± 18†	51 ± 11†	16 ± 5§	14 ± 4§
Patients without angiographic CAD	24	63 ± 13‡	56 ± 14‡	7 ± 1§	5 ± 1§

*p = 0.001.
†p = 0.01.
‡p = 0.04.
§p = 0.02.
FDI = flow decrease index.

CAD or apparent normal coronary arteries. The patient group without angiographic evidence of CAD may have developed diffuse small-vessel disease to an extent not detected by angiography.

PET is an advanced imaging technique that allows quantification of MBF (*11*) and metabolic status (*14–15*) of the myocardium noninvasively by tracing the kinetics of radiolabeled physiological substrates. Krivokapich et al. quantified absolute myocardial perfusion with PET using ^{13}N-ammonia in normal volunteers and subsequently reported using that technique to detect primary CAD (*16*). Demer et al. demonstrated the usefulness of PET using ^{13}N-ammonia and ^{82}Rb as tracers to assess the severity of primary CAD (*10*). Their results showed that PET measurements of MBF correlated with measurements of MBF by quantitative computerized angiography (*10*).

Several centers have evaluated the role of PET when examining heart transplant recipients. By using PET in a rat heterotopic heart transplant model, Hoff et al. demonstrated that ^{13}N-ammonia and ^{18}F-FDG can detect decreased myocardial perfusion and increased metabolic rate in cardiac allografts during acute rejection (*12*). Similarly, several groups have studied absolute myocardial perfusion using PET in heart transplant recipients with no angiographic evidence of CAD (*24–26*). Their results show that resting coronary blood flow was significantly higher in cardiac allografts compared to normal volunteers. Allograft vascular response to exercise (*24*) or dipyridamole stress (*25,26*) was preserved. Studies using Doppler flow velocity measurements at rest and following maximal vasodilation with papaverine also found that coronary blood flow reserve in transplant recipients was not significantly different from that of normal volunteers (*27*). In these studies, however, there were no sequential measurements over time. Nitenberg et al. have measured coronary blood flow reserve (ratio of peak papaverine to rest measurements) over time in cardiac transplant recipients using Doppler coronary flow velocity measurements (*28*). Although they found no difference in coronary blood flow reserve between normal volunteers and heart transplant patients with angiographically normal coronaries even late after transplantation, they did not measure absolute myocardial

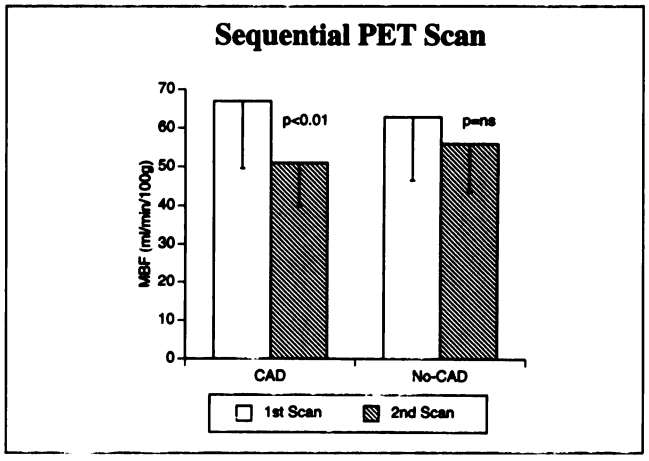

FIGURE 2. Changes in global MBF on sequential PET scans in the patients with and without angiographic evidence of CAD.

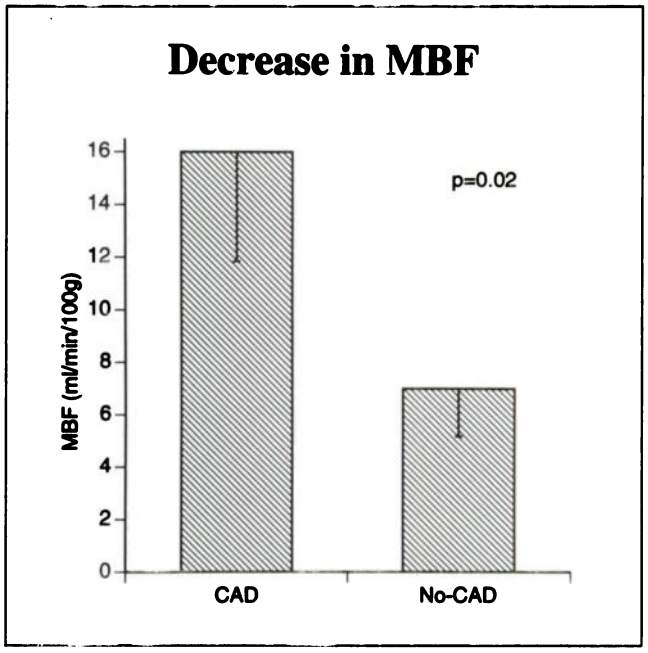

FIGURE 3. Decrease in MBF in the patients with and without angiographic evidence of CAD.

TABLE 3
Risk Factors in Patients with and without MBF Decrease by PET

Risk factors	MBF decrease by PET	No MBF decrease by PET	Student t-test
Sex	19 M/3 F	12 M/1 F	p= ns
Recipient age (yr)	44 ± 13	41 ± 17	p= ns
Donor age (yr)	26 ± 9	24 ± 10	p= ns
Ischemic time (min)	141 ± 44	123 ± 60	p= ns
HLA mismatch	4.6 ± 1.1	3.8 ± 1.2	p= ns
Rejection (Event free at 12 mo)	25 ± 9%	46 ± 8%	p=0.06
CMV infection (Event free at 12 mo)	35 ± 9%	45 ± 9%	p=0.07

blood flow. Since vascular intimal smooth muscle hyperplasia characterizes allograft CAD as opposed to calcification or fibrosis in primary CAD, the vessels in allograft CAD may be able to respond to vasodilators even if the resting MBF decreases. Studies using PET (*29*) and intracoronary Doppler flow velocity measurements (*30*) demonstrate that coronary vascular reserve is altered during acute allograft rejection.

We applied PET in heart transplant recipients in an attempt to assess accurately the progression of allograft CAD noninvasively. The hypothesis is that the development of progressive diffuse intimal thickening of the coronary arteries (allograft CAD) may result in a decrease in global MBF, detectable by measuring an index of absolute myocardial perfusion. The results demonstrate that a decrease in MBF was detected by PET in 63% (22/35) of patients who survived more than a year after transplantation. In contrast, only 31% (11/35) of patients had angiographic evidence of CAD. A decrease in MBF was more profound in patients with than those without angiographic evidence of CAD. The decrease in MBF over time in sequential scans appears to be more sensitive for diagnosing allograft CAD than angiography because the proportion of patients with a decrease in MBF (63%) reflects the true incidence of allograft CAD detected pathologically (*6*) more closely than the proportion of patients with angiographic evidence of CAD (31%).

Without an established gold standard for the detection of allograft CAD in vivo, it is possible that PET overestimated the prevalence of allograft CAD. Clinical and pathological studies, however, suggest that the PET findings may in fact be accurate. Johnson et al. reported the disparity between angiographic and postmortem findings, pointing out the insensitivity of coronary angiography in the detection of allograft CAD (*4*). A group of researchers recently described intravascular ultrasound findings consistent with the intimal thickening of allograft CAD that was not evident on coronary angiography (*8–9*). Based on these reports and clinical experience, PET data in this study may reflect the true incidence of allograft CAD more closely than the coronary angiographic findings. Correlation of PET with quantitative computerized angiography and intracoronary ultrasound is currently under investigation at our institute.

Sequential PET scans served to monitor changes in myocardial perfusion with time. The decrease in MBF observed in patients over a period of time is consistent with the known progressive nature of allograft CAD. There was no significant difference in the pattern of flow changes in different left ventricular segments, reflecting the diffuse and uniform nature of the disease process.

Mechanisms underlying CAD are not clear. Immunological injury is implied in the development of CAD as allograft rejection and the production of anti-donor HLA antibodies are associated with the development of CAD. We previously demonstrated that production of growth factors for vascular smooth muscle cells and expression of their receptors are increased significantly in cardiac allografts during rejection and are associated with CAD (*20–23*). Patients with significant MBF decrease had more rejection episodes and higher CMV infection rates compared to those without flow decrease; however, statistical significance was not achieved.

CONCLUSION

Our data suggest that allograft CAD can be detected noninvasively with serial PET studies. In the absence of a reference standard for detection of allograft CAD in vivo, it is difficult to determine the diagnostic sensitivity, specificity and predictive accuracy of PET. Our findings, however, indicate that PET is at least more sensitive than coronary angiography for the diagnosis of allograft CAD. PET therefore holds promise as a useful noninvasive modality for detecting the development and monitoring of allograft CAD.

REFERENCES

1. Uretsky BF, Murali S, Reddy PS, Rabin B. Development of coronary artery disease in cardiac transplant patients receiving immunosuppressive therapy with cyclosporine and prednisone. *Circulation* 1987;76:827–834.
2. Billingham ME. Cardiac transplant atherosclerosis. *Transplant Proc* 1987; 19(suppl 5):19–25.
3. Johnson DE, Gao SZ, Schroeder J, et al. The spectrum of coronary artery pathologic findings in human cardiac allografts [Abstract]. *J Heart Lung Transplant* 1989;8:349.
4. Gao SZ, Alderman EL, Schroeder JS, Silverman JF, Hunt SA. Accelerated coronary vascular disease in the heart transplant patients: coronary arteriographic findings. *J Am Coll Cardiol* 1988;12:334–340.
5. O'Neil BJ, Pflugfelder PW, Singh NR, Menkis AH, McKenzie FN, Kostuk WJ. Frequency of angiographic detection and quantitative assessment of

coronary arterial disease one and three years after cardiac transplantation. *Am J Cardiol* 1989;63:1221–1226.
6. Russell ME, Fujita M, Masek MA, Rowan RA, Billingham ME. Cardiac graft vascular disease, nonselective involvement of large and small vessels. *Transplantation* 1993;56:762–763.
7. Gao SZ, Alderman EL, Schroeder JS, Hunt SA, Wiederho DV, Stinson EB. Progressive coronary luminal narrowing after cardiac transplantation. *Circulation* 1990;82(suppl 4):IV269–IV275.
8. Valantine H, Pinto FJ, St Goar FG, Alderman EL, Popp RL. Intracoronary ultrasound imaging in heart transplant recipients: the Stanford experience. *J Heart Lung Transplant* 1992;11:S60–S64.
9. St Goar FG, Pinto FG, Alderman EL, et al. Intracoronary ultrasound in cardiac transplant recipients. In vivo evidence of angiographically silent intimal thickening. *Circulation* 1992;85:979–987.
10. Demer LL, Gould KL, Goldstein RA, et al. Assessment of coronary artery disease severity by positron emission tomography. *Circulation* 1989;79:825–835.
11. Kuhle WG, Porenta G, Huang SC, et al. Quantification of regional myocardial blood flow using ^{13}N-ammonia and reoriented dynamic positron emission tomographic imaging. *Circulation* 1992;86:1004–1017.
12. Hoff SJ, Stewart JR, Frist WH, et al. Noninvasive detection of heart transplant rejection with positron emission scintigraphy. *Ann Thorac Surg* 1992;53:572–577.
13. Frist WH, Stewart JR, Merrill WH, et al. Positron emission tomography (PET) noninvasively detects diminishing coronary blood flow after heart transplant [Abstract]. *J Heart Lung Transplant* 1993;10:174.
14. Sokoloff L, Reivich M, Kennedy C, et al. The (^{14}C)deoxyglucose method for the measurement of local cerebral glucose utilization: theory, procedure and normal values in the conscious and anesthetized albino rat. *J Neurochem* 1977;28:897–916.
15. Ratib O, Phelps ME, Huang SC, Henze E, Selin CE, Schelbert HR. Positron emission tomography with deoxyglucose for estimating local myocardial glucose metabolism. *J Nucl Med* 1982;23:577–586.
16. Krivokapich J, Smith GT, Huang SC. Nitrogen-13-ammonia myocardial imaging at rest and with exercise in normal volunteers. *Circulation* 1989; 80:1328–1337.
17. Votaw JR, Delbeke D, Woods R, Holburn G, Sandler MP. Comparison of different techniques for calculating myocardial blood flow [Abstract]. *J Nucl Med* 1993;34:86P.
18. Rosenpire KC, Schwaiger M, Manger TJ, Hutchins GD, Sutirik A, Kuhl DE. Metabolic fate of ^{13}N-ammonia in human and canine blood. *J Nucl Med* 1990;31:163–167.
19. Uren NG, Melin JA, De Bruyne B, Wijns W, Baudhiun T, Camici PG. Relation between myocardial blood flow and the severity of coronary artery stenosis. *N Engl J Med* 1994;330:1782–1788.
20. Zhao XM, Yeoh TK, Hiebert M, Frist WH, Miller GG. The expression of acidic fibroblast growth factor (heparin binding growth factor-1) and cytokine genes in human cardiac allografts and T cells. *Transplantation* 1993; 56:1177–1182.
21. Zhao XM, Frist WH, Yeoh TK, Miller GG. Confirmation of alternative spliced platelet-derived growth factor-A chain and correlation with expression of PDGF receptor alpha in human cardiac allografts. *Transplantation* 1994:in press.
22. Zhao XM, Frist WH, Yeoh TK, Miller GG. Modification of mRNA splicing of fibroblast growth factor receptors in human cardiac allografts. *J Clin Invest* 1994;94:992–1003.
23. Zhao XM, Yeoh TK, Frist WH, Miller GG. Induction of fibroblast growth factor and platelet-derived growth factor gene expression in human cardiac allografts. *Circulation* 1994;90:677–685.
24. Krivokapich J, Stevenson L, Kobashigawa J, Huang SC, Schelbert HR. Quantification of absolute myocardial perfusion at rest and during exercise with positron emission tomography after human cardiac transplantation. *J Am Coll Cardiol* 1991;18:512–517.
25. Rechavia E, Araujo L, De Silva R, et al. Dipyridamole vasodilator response after human orthotopic heart transplantation: quantification by oxygen-15-labeled water and positron emission tomography. *J Am Coll Cardiol* 1992; 19:100–106.
26. Senneff MJ, Hartman J, Sobel BE, Geltman EM, Bergmann SR. Persistence of coronary vasodilator responsivity after cardiac transplantation. *Am J Cardiol* 1993;71:333–338.
27. McGinn AL, Wilson RF, Olivari MT, Homans DC, White CW. Coronary vasodilator reserve after orthotopic cardiac transplantation. *Circulation* 1988;78:1200–1209.
28. Nitenberg A, Aptecar E, Benvenuti C, Benhaiem N, Tavolaro O, Loisance D, Cachera JP. Effects of time and previous acute rejection episodes on coronary vascular reserve in human heart transplant recipients. *J Am Coll Cardiol* 1992;20:1333–1338.
29. Chan SY, Kobashigawa J, Stevenson LW, et al. Basal myocardial blood flow is increased by maximal flow is decreased during acute cardiac transplant rejection: a non-invasive quantitative study [Abstract]. *J Am Coll Cardiol* 1991;17:58A.
30. Nitenberg A, Tavolaro O, Benvenuti C, et al. Recovery of a normal coronary vascular reserve after rejection therapy in acute human cardiac allograft rejection. *Circulation* 1990;81:1312–1318.