Lumen Loss in Transplant Coronary Artery Disease Is a Biphasic Process Involving Early Intimal Thickening and Late Constrictive Remodeling
Results From a 5-Year Serial Intravascular Ultrasound Study

Hiroshi Tsutsui, MD; Khaled M. Ziada, MD; Paul Schoenhagen, MD; Atila Iyisoy, MD; William A. Magyar, BS; Timothy D. Crowe, BS; Jon D. Klingensmith, MS; D. Geoffrey Vince, PhD; Gustavo Rincon, MD; Robert E. Hobbs, MD; Masakazu Yamagishi, MD; Steven E. Nissen, MD; E. Murat Tuzcu, MD

Background—Coronary artery disease is the major cause of late cardiac allograft failure. However, few data exist regarding the natural history of changes in intimal and external elastic membrane (EEM) areas after heart transplantation.

Methods and Results—In 38 transplant recipients, serial intravascular ultrasound examinations were performed 3.7±2.2 weeks after transplantation and annually thereafter for 5 years. In 59 coronary arteries, we compared 135 matched segments among serial studies. In each segment, intravascular ultrasound images were digitized at 1-mm intervals, and mean values of EEM and lumen and intimal areas were analyzed. In the first year after transplantation, the intimal area increased significantly from 1.8±1.6 to 3.0±2.1 mm² (P<0.001). Subsequently, the annual increase in intimal area decreased. EEM area did not change during the first year; however, between years 1 and 3, significant expansion of EEM area occurred (15.4±4.6 to 17.2±5.4 mm², P<0.001). Thereafter, EEM area decreased significantly from 17.2±5.4 mm² (year 3) to 15.1±4.9 mm² (year 5, P<0.01). Different mechanisms of lumen loss were observed during 2 phases after transplantation: early lumen loss primarily caused by intimal thickening and late lumen loss caused by EEM area constriction.

Conclusions—This serial ultrasound study revealed that most of the intimal thickening occurred during the first year after heart transplantation. Changes in the EEM area showed a biphasic response, consisting of early expansion and late constriction. Thus, different mechanisms of lumen loss were observed during the early and late phases after transplantation. (Circulation. 2001;104:653-657.)

Key Words: transplantation ■ ultrasonics ■ cardiovascular diseases

Transplant coronary artery disease (CAD) is the principal cause of late death in heart transplant recipients.1–3 Previous studies have demonstrated the advantages of intravascular ultrasound (IVUS) imaging in identifying this entity.4–6 IVUS imaging quantifies both intimal thickening and changes in external elastic membrane (EEM) area (arterial remodeling) in transplant CAD, and several studies have emphasized that lumen loss is caused not only by intimal thickening but also by changes in EEM area.7–10 At present, there are no long-term, systematic follow-up data regarding the relative importance of intimal thickening and EEM area changes in transplant CAD.

The objectives of this study were to assess the time course of progressive intimal thickening, EEM area changes, and effects of these changes on lumen loss in patients over a 5-year period after heart transplantation.

Methods

Patient Population

The study population consisted of heart transplant recipients who underwent IVUS imaging early after transplantation and annually thereafter. Between January 1993 and April 1996, 133 heart transplant recipients underwent IVUS examinations within 8 weeks after transplantation. Of these patients, complete annual IVUS studies for at least 4 years were available in 44 recipients. We excluded 6 patients because of imaging artifacts in the IVUS recordings. The study protocol was approved by the Institutional Review Board of the Cleveland Clinic Foundation.

Coronary Angiography and IVUS Examination

Coronary angiography and IVUS examination were performed with the use of standard techniques as previously reported.6 Multiple angiographic views were obtained with 7F coronary guiding catheters. After anticoagulation with heparin and intracoronary administration of 100 to 200 µg nitroglycerin, IVUS imaging was performed...
with 30-MHz ultrasound transducers (3.5F Soni cath: Boston Scientific/3.2F Ultracross; CVIS) interfaced with dedicated scanners (Hewlett-Packard/CVIS-Boston Scientific). The IVUS catheter was inserted into the distal portion of the coronary artery over a 0.014-in angioplasty guide wire, and the catheter location was recorded with cineangiography. The ultrasound catheter was withdrawn slowly and steadily with manual pullback. After 1998, a motorized pullback system (0.5 mm/s) was used during IVUS imaging. IVUS images were recorded on S-VHS tape with the use of voice annotation.

**IVUS Analysis**

Coronary segments were identified by using major coronary branches as starting points and end points. The length of the segment was assessed as the distance between these two landmarks measured from the last annual examination. In 19 patients (73 segments), the latest IVUS examination was recorded with motorized pullback, and we obtained the length of segments by using time scales of the motorized pullback. In the remaining 19 patients (62 segments), in whom only manual pullback was used, the latest cineangiography was used for length measurements of the coronary segments. In the least foreshortened angiographic view, the segment length was measured by a computer with NIH software. The length of the segment defined the number of IVUS still frames (at 1-mm intervals), which were selected from the matched coronary segments in every year (Figure 1). With the use of a semiautomated tracing system, lumen area and EEM area were measured in each still image by one investigator who was blinded to the clinical information. Intimal area (EEM area–lumen area: theoretically including intima and media) and percent area reduction [(Intimal area/EEM area)×100] were calculated. A mean value of each variable was calculated for every matched coronary segment and compared among serial examinations.

To assess the reproducibility of this method, we performed subsequent motorized and manual pullbacks of the IVUS catheter during the same IVUS examination. In 10 coronary segments, mean values of the EEM and lumen areas were calculated on the basis of these two IVUS recordings. The percent errors between the two analyses were 0.71±2.84% for the EEM area and 0.68±3.73% for the lumen area. The correlation coefficient was 0.99 for each comparison. We also reanalyzed the mean values of the EEM and lumen areas in 10 coronary segments after 4 weeks (repeat frame selections and repeat measurements), based on the same IVUS recordings. The percent errors were 1.11±1.55% for the EEM area and 0.01±2.27% for the lumen area. The correlation coefficients were 0.99 for both the comparisons.

**Data Analysis**

Normally distributed data were presented as mean±1 SD. Comparison with average values of serial IVUS measurements were determined by mixed modeling for repeated measures with Tukey-Kramer adjustments for multiple comparisons. A value of *P*<0.05 was considered to be statistically significant.

**Results**

**Characteristics of Patients and Coronary Arteries**

Complete annual IVUS examinations for at least 4 years after transplantation were available in 38 transplant recipients (28 men; mean age, 52±8.5 years). The first IVUS examinations were performed 3.7±2.2 weeks after transplantation. In 17 of these patients, serial IVUS recordings were available over a period of 5 years. Indications for heart transplantation were idiopathic dilated cardiomyopathy (*n*=17) and ischemic heart disease (*n*=21). The mean donor age was 29±12 years, and 26 donors were men. We analyzed annual images of 135 matched coronary segments in 59 coronary arteries (left anterior descending, 28; left circumflex, 16; right coronary, 15). The average length of segments was 12.5±5.4 mm.

**Serial Changes in Intimal Area**

During the first 2 years after heart transplantation, the average intimal area increased significantly from 1.8±1.6 mm² (baseline) to 3.0±2.1 mm² (year 1) and to 3.8±2.5 mm² (year 2) (*P*<0.001, respectively) (Figure 2). After year 2, there was no significant increase in intimal area. Yearly increases in intimal area accounted for 52% (year 1), 34% (year 2), and 14% (year 3) of the total intimal thickening during follow-up. The rate of increase in the intimal area decreased each year.

**Serial Changes in EEM Area**

The time course of EEM area changes is shown in Figure 3. In the first year after heart transplantation, the average EEM area did not change significantly (15.8±4.7 mm² to 15.4±4.6 mm²). However, between year 1 and year 3, the EEM area expanded significantly from 15.4±4.6 mm² to 17.2±5.4 mm² (*P*<0.001). Thereafter, the EEM area decreased from 17.2±5.4 mm² (year 3) to 15.1±4.9 mm² (year 5, *P*=0.01). After no significant change within the first year,
The present study demonstrated that (1) intimal thickening occurred early after heart transplantation, mainly in year 1 and less in years 2; (2) the average EEM area did not change within the first year but then showed a biphasic response, consisting of early expansion and late constriction. (3) Resultant lumen losses were caused by different mechanisms during the two phases after transplantation: Early lumen loss was primarily caused by intimal thickening and late lumen loss was caused by EEM area constriction. These mechanisms of lumen loss suggest that the pathophysiology of transplant CAD may be different in the early and late phases after heart transplantation.

Although several investigators have reported different aspects of transplant CAD with the use of IVUS imaging, there are some unique features to this study. To our knowledge, this study is the first report of a long-term, serial IVUS evaluation in transplant CAD up to 5 years after heart transplantation. Importantly, initial IVUS imaging was performed early after heart transplantation, which allowed detection and follow-up of transplant CAD from the onset. Systematic annual imaging allowed changes in EEM area and intimal thickening to be analyzed in a time perspective. We analyzed average data of multiple still images in each matched coronary segment rather than single matched frames. This approach is probably more representative of diffuse changes in transplant CAD.

**Serial Changes in EEM Area**

In the first year after transplantation, the average lumen area decreased significantly from $13.9 \pm 3.7 \text{ mm}^2$ (baseline) to $12.4 \pm 3.8 \text{ mm}^2$ (year 1, $P < 0.001$). Between year 1 and year 3, the lumen area remained unchanged ($12.7 \pm 4.1 \text{ mm}^2$ [year 2] and $13.0 \pm 4.0 \text{ mm}^2$ [year 3]). However, in the last 2 years of follow-up, the lumen area decreased significantly from $13.0 \pm 4.0 \text{ mm}^2$ (year 3) to $11.4 \pm 3.9 \text{ mm}^2$ (year 5, $P = 0.007$) (Figure 4). Within the 5-year follow-up interval, significant lumen losses were observed during two phases, the first year and the last 2 years of the follow-up. However, the mechanisms of lumen loss were different in these phases (Figure 5). In the initial year after transplantation, 79% of the lumen loss was caused by intimal thickening and only 21% was related to constriction of EEM area. On the other hand, during the last 2 years of the follow-up period, the decrease of EEM area was exclusively responsible for lumen loss (Figure 6).

**Intimal Thickening**

Our finding that most intimal thickening occurs in the first year is consistent with previous studies. Rickenbacher et al demonstrated that intimal thickening appeared early after cardiac transplantation, mostly in year 1. Recently, Kobashigawa et al reported similar findings about intimal thickening in a serial IVUS study of transplant CAD over a period of 3 years. These findings imply that potential interventions to ameliorate intimal thickening of transplant CAD should be started early after transplantation.

**Changes in EEM Area**

Several investigators have described EEM area changes in transplant CAD by using IVUS imaging. Lim et al compared the findings of 2 consecutive IVUS examinations in 75 patients performed at varying intervals after heart transplantation. These investigators showed that expansion of EEM area occurred in relative early phase (2.0 ± 0.3 years) after transplantation, whereas EEM area constriction was observed later (3.2 ± 0.5 years). On the other hand, Pethig et al demonstrated that EEM area decreased within the first year after transplantation and increased thereafter. Our findings from serial observations provide an explanation for these seemingly contradictory results. During the first year after transplantation, we detected lack of remodeling or slight decrease in the EEM area, which supports the results of Pethig et al. The subsequent biphasic response in EEM area, early expansion (between years 1 and 3) and late constriction (between years 3 and 5), is consistent with the data by Lim et al.

We observed an increase in EEM area associated with intimal thickening between year 1 and year 3 after transplantation. This response in EEM area is similar to compensatory positive remodeling in early atherosclerosis of native coronary arteries as initially described by Glagov et al and subsequently confirmed by several IVUS studies. The pathophysiology of coronary remodeling in native coronary arteries is not completely understood; however, endothelial cells and the extracellular matrix appear to play important roles. In coronary arteries after transplantation, endothelial dysfunction and coronary vasculitis were reported and probably influence coronary remodeling. In our study, the average EEM area did not increase despite marked intimal
thickening during the first year after transplantation. Allograft responses to intense immunologic activity early after transplantation may limit the remodeling process.25 The mechanisms of remodeling in transplant CAD could be more complicated than that in native coronary arteries.

Our findings of late EEM area constriction has also been reported in previous IVUS studies of transplant CAD.7,10 In a necropsy study of transplant CAD, Arbustini and Roberts26 described thick, dense circumferential adventitial fibrosis with occasional clusters of inflammatory cells. They also showed distorted epicardial coronary arteries caused by constriction from adventitial fibrosis. Thus, a chronic inflammatory process with subsequent fibrosis may explain late EEM area constriction. However, the exact mechanisms of EEM area changes in the disease process remains unclear.

Mechanisms of Lumen Loss
The systematic year-to-year comparison revealed time-dependent changes in intimal and EEM areas. The interactions between these changes explain the different mechanisms of lumen loss in the early and late phases after heart transplantation. Pethig et al8 analyzed mechanisms of lumen loss by comparing reference and lesion sites and by comparing serial changes in two IVUS examinations.9 The investigators concluded that the contribution of EEM area change to lumen loss is greater than that of intimal thickening. Our findings show different time courses in intimal thickening and EEM area changes after heart transplantation. Therefore, it may be difficult to determine the mechanisms of lumen loss in transplant CAD without systematic serial IVUS examinations.

Limitations
The study population is relatively small. However, obtaining complete serial IVUS examinations for 5 years without image artifacts is extremely difficult. The study population has an inherent selection bias toward less severe disease because patients with aggressive transplant CAD may not have survived 5 years and those with angiographically severe CAD were not imaged with IVUS. Other methodological limitations include the use of two types of IVUS systems during follow-up. This device change may have affected our measurements. In addition, most of the early IVUS imaging was performed with a steady manual pullback of the IVUS catheter. We cannot exclude the possibility that less uniform pullback speed of manual pullbacks has affected our analyses. Volumetric analysis with motorized pullbacks now is the most suitable approach to compare serial changes on IVUS examinations.9 However, motorized pullbacks were not available during the early period of this study.

Intima without media cannot be measured accurately with the use of IVUS. Therefore, the definition of “intima plus media” area is used in IVUS measurements of CAD. However, pathological studies of transplant vasculopathy show that changes in “intima plus media” area consist mainly of intimal proliferation.22,24,26 Therefore, we used terms of “intimal area” and “intimal thickening” in this article.

Conclusions
Using serial IVUS imaging in transplant coronary arteries, we demonstrated that most intimal thickening occurred within the first year after heart transplantation. The EEM area showed a biphasic response with early expansion followed by late constriction during follow-up. As a consequence of these changes, significant lumen loss was observed in the initial year and during the last 2 years of follow-up. However, the mechanisms of lumen loss were different. Early lumen loss was caused primarily by intimal thickening and late lumen loss occurred because of constriction of the EEM area.
Acknowledgments
Dr Tsutsui is supported partially by a research grant from the Japanese Association for the Advancement of Medical Equipment, affiliated with the Ministry of Health and Welfare of Japan, Tokyo. We thank Michelle Secic, MS, for statistical analysis.

References
Lumen Loss in Transplant Coronary Artery Disease Is a Biphasic Process Involving Early Intimal Thickening and Late Constrictive Remodeling: Results From a 5-Year Serial Intravascular Ultrasound Study


Circulation. 2001;104:653-657
doi: 10.1161/hc3101.093867

Circulation is published by the American Heart Association, 7272 Greenville Avenue, Dallas, TX 75231
Copyright © 2001 American Heart Association, Inc. All rights reserved.
Print ISSN: 0009-7322. Online ISSN: 1524-4539

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circ.ahajournals.org/content/104/6/653