Regions of low endothelial shear stress are the sites where coronary plaque progresses and vascular remodelling occurs in humans: an in vivo serial study

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Aims We performed serial intracoronary studies of patients with stable coronary artery disease (CAD) to investigate the relationships among baseline endothelial shear stress (ESS), CAD progression, and vascular remodelling. Local haemodynamic factors are critical determinants of plaque progression, vascular remodelling, and clinical CAD manifestations.

Methods and results The 3-D anatomy of coronary arteries with lumen obstruction, 50% was determined by fusing intracoronary ultrasound and angiographic images in 13 patients at baseline and 8 ± 2 months later. Cross-sectional area of plaque, lumen, and external elastic membrane (EEM), and coronary flow were measured. Local ESS was calculated. Subsegments with similar ESS were categorized based on low (< 12 dynes/cm²) and moderate/higher ESS (≥ 12 dynes/cm²). There were 47 subsegments of similar baseline ESS: nine with low ESS and 38 with moderate/higher ESS. Median subsegment length was 6.9 mm (25th–75th percentiles = 4.2–12.0), and median area of similar ESS of 52.6 mm² (25th–75th percentiles = 26.9–88.0). Subsegments with low ESS exhibited plaque progression when compared with subsegments with moderate/higher ESS (33.3% vs. 7.9%, respectively, P = 0.009 adjusted for clustering of lesions within patients) and constrictive remodelling (44.0% vs. 5.3%, respectively, P = 0.16 adjusted for clustering of lesions within patients). Expansive remodelling occurred with similar frequency in subsegments with low vs. moderate/higher baseline ESS.

Conclusion Plaque progresses in subsegments with low ESS, associated with either constrictive or expansive remodelling. Different mechanisms are likely responsible for expansive remodelling in different local vascular environments. Early in vivo identification of arterial subsegments likely to develop high-risk plaque characteristics may allow for selective interventions to avoid adverse cardiac outcomes.

Introduction

Progression of coronary artery disease (CAD) is related both to the growth of atherosclerotic plaque and to either constrictive or expansive vascular remodelling.1–6 The Glagov concept of obstructive CAD7 assumes that the external elastic membrane (EEM) only expansively remodels as plaque progresses and that lumen encroachment occurs when plaque progression outstrips the EEM’s ability to expand. Vascular remodelling responses, however, are more complex: although up to 60% of lesions exhibit ‘compensatory’ outward remodelling, a substantial proportion exhibit ‘excessive’ remodelling (EEM area increased in excess of the increase in plaque area), and a substantial proportion remodel ‘inadequately’ or constrictively with resulting lumen narrowing.8,9 The clinical significance of these remodelling patterns is critical since plaques within expansively remodelled segments are associated with plaque vulnerability and presentation with an acute coronary syndrome, whereas plaques within constrictively remodelled segments are more fibrous and associated with a stable clinical presentation.10–13

Local haemodynamic factors, especially low endothelial shear stress (ESS), are important determinants of atherosclerotic plaque progression and vascular remodelling.14–18 If it were possible to identify in vivo early plaques that were likely to develop characteristics of either vulnerable plaque or lumen obstruction, then selective interventions could be applied to these areas to avoid subsequent cardiac events. The purpose of this pilot study was to identify arterial subsegments with predominant low vs.
methodology to investigate the effects of different ESS environments on plaque and wall characteristics ≥ 6 months later.

Methods
Subjects were recruited from patients undergoing elective cardiac catheterization for clinical indications. Subjects were eligible if they underwent coronary stent deployment and had another coronary artery with lumen obstruction ≤ 50%. Exclusion criteria included ejection fraction < 40%, serum creatinine > 1.5 mg/dL, 3-vessel CAD, or valvular disease. Intracoronary vascular profiling was performed on the artery with < 50% obstruction after administration of parenteral nitroglycerin or verapamil. The population consisted of 13 patients: six patients enrolled in a previous pilot study, and an additional seven patients using the same protocol. Patients were followed for ≥ 6 months and then returned for repeat vascular profiling. The Human Subjects Committee of Brigham & Women’s Hospital approved the study. Each patient provided written informed consent. This report presents the natural history of the unstented, minimally narrowed coronary arteries.

Intracoronary vascular profiling
Our methods have been previously described in detail. In brief, the 3-D anatomy of the artery was reconstructed from IVUS images and biplane coronary angiography. IVUS (Atlantis Catheter, Boston Scientific, Natick, MA, USA) was performed with mechanized pullback at 0.5 mm/s. The arterial lumen and outer vessel wall (EEM) were segmented from digitized end-diastolic IVUS frames. The physical 3-D path of the IVUS transducer during pullback was reconstructed using the corresponding biplane angiographic projections. The 3-D reconstructed catheter core served as the stem on which to rebuild the 3-D geometry. The 3-D position of each ECG-gated IVUS frame was determined from the pullback speed and the reconstructed trajectory of catheter pullback, and its spatial orientation was matched with the angiographic appearance of the lumen. The boundary points of each frame were connected by spline curves to rebuild the lumen and the EEM geometry in 3-D space, using the lumen and EEM boundary points, respectively. Plaque was defined as the region between the vessel outer wall and the lumen. A structured grid utilizing a body-fitted coordinate system was employed to represent the lumen volume. Coronary blood flow for the arterial section being studied was calculated directly from the time required for the volume of blood contained within the section to be displaced by radio-opaque material during a contrast injection, as previously described.

Detailed intravascular flow characteristics were obtained by numerically solving the transport equations governing the conservation of mass and momentum. Shear stress at the lumen surface of the artery was calculated as the product of viscosity (calculated from the measured hematocrit) and the gradient of blood velocity at the wall.

The processes of data acquisition and data analysis are highly reproducible.

Identification of subsegments based on baseline ESS
ESS was mapped on the reconstructed lumen surface, regions of similar ESS were identified, and these regions were classified into two ESS categories at baseline: 'low ESS' (< 12 dynes/cm²), and 'moderate/higher ESS' (≥ 12 dynes/cm²).

Coronary artery 'subsegments of interest' were created from the reconstructed coronary arteries and categorized on the basis of the predominant ESS. This subsegment approach was chosen so that appropriate portions of the artery could be identified for consideration of pre-emptive percutaneous intervention. To be considered in the analysis, the subsegment was required to have a surface area region of similar ESS that was ≥ 5 mm² and length ≥ 3 mm. The region of homogeneous ESS was required to constitute ≥ 30% of the subsegment surface area and mean ESS in the entire subsegment was required to be within the range of values of the respective category. If two regions of similar ESS categories were closer than 1 mm from each other, they were combined and considered as one region. Each subsegment of interest was defined from the margins of the similar ESS regions (Figure 1).

The measurements made at baseline were compared with those at follow-up by matching the regions using fiducial sites based on IVUS-derived and angiography-confirmed anatomical landmarks.

Statistical analysis
Changes in plaque, EEM, and lumen cross-sectional areas in each subsegment were categorized as increased, decreased, or none,
where change was defined as a difference at follow-up from baseline that exceeded $\pm 3$ standard deviations of previously determined measurement repeatability (standard deviation of difference between repeated measurements for plaque, EEM, and lumen cross-sectional areas 0.4, 0.6, and 0.6 mm$^2$, respectively). 21 Moderate and higher ESS subsegments were combined because our hypothesis was that low ESS was the important determinant of pathobiologic changes. The association between ESS category and changes in plaque, EEM, and lumen cross-sectional area was assessed with ordered logistic regression. Ordered logistic regression was also employed to determine the association between plaque growth categorized as yes or no and EEM cross-sectional area. Standard errors of the regression coefficients were adjusted for the clustering of lesions within patients with the Huber White Sandwich Estimator. We also investigated the relationship of change in plaque, EEM, and lumen and baseline ESS using a continuous scale. The standard errors of the regression coefficients were adjusted for intra-group correlation with mixed effects models (Stata version 9). A similar approach was used to determine the bivariate relationships between three separate pairs of the continuous measures of change in plaque, EEM, and lumen cross-sectional areas. All statistical tests were two-sided with $\alpha = 0.017$ in order to adjust for the multiple comparisons of data related to ESS and cross-sectional areas.

Results

Patient and coronary artery subsegment characteristics

Twenty-five subjects with stable CAD underwent initial catheterization and vascular profiling between May 2000 and March 2003. Eight of these subjects had vascular profiling only of a stented artery and were not suitable for inclusion in this natural history study. Of the 17 subjects with evaluation of a non-stented artery with minimal lumen obstruction, 13 underwent follow-up study a mean of 8 ± 2 months later. Three subjects refused follow-up catheterization, and one subject developed an acute coronary syndrome requiring revascularization and was not appropriate for follow-up study. Among the 13 subjects with serial studies (Table 1), 47 subsegments were identified from the baseline study with a predominant level of ESS: one subject contributed one subsegment, three subjects contributed two, two subjects three, five subjects four, one subject six, and one subject eight subsegments. Nine subsegments were classified as low ESS and 38 as moderate/higher ESS. No patient had more than one subsegment with low ESS. These homogeneous artery subsegments had a median length of 6.9 mm, with 25th–75th percentiles = 4.2–12.0, median area of similar ESS of 52.6 mm$^2$, with 25th–75th percentiles = 26.9–88.0, and each artery contained a mean of 3.6 ± 1.1 subsegments. Five patients (38%) were diabetic and contributed 10 subsegments to the analysis: two with low and eight with moderate/higher ESS.

Changes in plaque area, EEM area, and lumen area

Figure 2 illustrates the categorical changes at follow-up in the subsegments with baseline low ESS vs. those with baseline moderate/higher ESS. At follow-up, plaque area progressed in three of nine (33.3%) of low ESS subsegments and three of 38 (7.9%) of moderate/higher ESS subsegments. The finding for plaque area was statistically significant, even after raising the alpha level from 0.05 to 0.017 to account for multiple comparisons ($P = 0.009$ by ordered logistic regression). The patterns of vascular remodelling (EEM changes) and lumen changes also differed according to baseline ESS. Constrictive remodelling occurred almost exclusively in subsegments with low ESS (four of nine subsegments, 44%) when compared with subsegments with moderate/higher ESS (two of 38 subsegments, 5.3%). Expansive remodelling occurred in a similar proportion of low and moderate/higher ESS subsegments (two of nine subsegments, 22% vs. 10 of 38 subsegments, 26.3%, respectively) ($P = 0.16$ by ordered logistic regression). Lumen area changes paralleled changes in EEM area in each ESS group ($P = 0.17$ by ordered logistic regression). These findings for EEM and lumen area changes were consistent with study hypotheses but were not statistically significant. An alternative analysis of ESS and changes in plaque, EEM, and lumen areas measured continuously supported the results presented in Figure 2.

There were insufficient numbers of subsegments from diabetic patients to perform meaningful statistical analyses based on diabetic status.

Correlation between changes in plaque area, EEM area, and lumen area

At baseline, the average cross-sectional area for plaque, EEM, and lumen was $7.9 \pm 4.1$, $17.3 \pm 6.8$, and $9.4 \pm 4.3$ mm$^2$, respectively. There was a high correlation between change in EEM area and change in lumen area ($r = 0.92$, $P < 0.0001$ by linear regression), but no association between the change in plaque area and change in lumen area ($r = 0.08$, $P = 0.62$, by linear regression) (Figure 3). Every subsegment in which plaque progressed exhibited a change in EEM area, and there was a moderate positive correlation between

### Table 1: Patient baseline characteristics

| Age (years) (mean ± SD) | 61 ± 11 |
| Gender (n, % male) | 9 (69%) |
| Race (n, % white) | 12 (92%) |
| History of | |
| Hypertension (n, %) | 11 (85%) |
| Diabetes mellitus (n, %) | 5 (38%) |
| Cigarette smoking (n, %) | 4 (31%) |
| Hyperlipidaemia (n, %) | 13 (100%) |
| Myocardial infarction (n, %) | 4 (31%) |
| Angina pectoris (n, %) | 12 (92%) |
| Medications | |
| Aspirin (n, %) | 13 (100%) |
| β-blockers (n, %) | 13 (100%) |
| Calcium channel blockers (n, %) | 1 (7%) |
| Long-acting nitrates (n, %) | 4 (31%) |
| Statins (n, %) | 13 (100%) |
| ACE inhibitors (n, %) | 5 (38%) |
| Blood pressure (mmHg) (mean ± SD) | 129 ± 11/75 ± 8 |
| Heart rate (b.p.m) (mean ± SD) | 65 ± 7 |
| Fasting lipid values | |
| Total cholesterol (mg/dL) (mean ± SD) | 147 ± 28 |
| Triglycerides (mg/dL) (mean ± SD) | 148 ± 61 |
| HDL (mg/dL) (mean ± SD) | 35 ± 10 |
| LDL (mg/dL) (mean ± SD) | 84 ± 23 |
| Coronary artery investigated | |
| Left anterior descending | 2 |
| Circumflex | 4 |
| Right coronary artery | 7 |
Atherosclerosis and arterial wall behaviour are highly complex and depend on plaque growth and the arterial remodelling responses. Although vascular behaviour is dependent on both genetic and systemic factors, the majority of pathobiologic manifestations of atherosclerosis are highly focal, reflecting the critical role of local endothelial behaviour. Our serial in vivo observations indicate that plaque progression in minimally diseased coronary artery subsegments occurs almost exclusively in areas of low ESS, and may be accompanied by either expansive or constrictive remodelling. The dual direction of vascular remodelling responses to plaque growth in low ESS areas is reflected in the moderate relationship between plaque growth and change in EEM ($r = 0.47$). Our first pilot study, which involved only six analysable subjects, served as a proof-of-concept that we could measure ESS in vivo, and that such measurements could predict subsequent plaque progression. In that study, we calculated ESS along the course of a coronary artery utilizing extremely small endothelial ‘patches’, each about 250 x 300 microns in size, and we analysed the arterial morphology underneath each patch, like an ‘ice pick’ involving the endothelial patch, the plaque, and the outer vessel wall. The patches were grouped into regions of like shear stress and each region was placed in one of six categories, according to its range of shear stress values. In that analysis, each arterial subsegment could contain multiple regions of similar shear stress, each of a different value. Each region was independently evaluated as a determinant for the development of plaque progression and vascular remodelling. In the current analysis, we more than doubled the sample size of the pilot study by including an additional seven analysable subjects and re-analyzing all data from the 13 subjects to address a much more refined and clinically relevant approach to evaluating CAD. In this analysis, we identify areas of similar ESS along the course of each artery at baseline to create a subsegment of interest with only one predominant ESS value, and each subsegment constitutes the unit of measure to investigate subsequent vascular behaviour.

The change in plaque area and change in EEM area ($r = 0.47$, $P = 0.007$).

Discussion

Atherosclerosis and arterial wall behaviour are highly complex and depend on plaque growth and the arterial remodelling responses. Although vascular behaviour is dependent on both genetic and systemic factors, the majority of pathobiologic manifestations of atherosclerosis are highly focal, reflecting the critical role of local endothelial behaviour. Our serial in vivo observations indicate that plaque progression in minimally diseased coronary artery subsegments occurs almost exclusively in areas of low ESS, and may be accompanied by either expansive or constrictive remodelling. The dual direction of vascular remodelling responses to plaque growth in low ESS areas is reflected in the moderate relationship between plaque growth and change in EEM ($r = 0.47$).

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here, is that this approach is exactly what would be used to evaluate and treat early, high-risk portions of coronary arteries in the cardiac catheterization laboratory.

Clinical importance of remodelling responses
Atherosclerotic plaques in expansively remodelled arterial segments exhibit characteristics of inflammation and are more likely to present as an acute coronary syndrome. Plaques in constrictively remodelled vascular subsegments are more fibrotic and calcified, and are more likely to present with a stable pattern of coronary disease.

Constrictive remodelling
Constrictive remodelling occurred in four (44%) of low ESS subsegments, but infrequently in subsegments with moderate or higher ESS. Of the four low ESS subsegments with constrictive remodelling, one was associated with plaque growth, and three constricted without plaque growth. Von Birgelen et al. also observed that constrictive remodelling occurred in the setting of an increase, decrease, or no change in plaque area.

Expansive remodelling
Expansive remodelling occurred in each ESS category, but likely occurred for different reasons in different local environments: in areas of active plaque progression (i.e. low baseline ESS) expansive remodelling is likely compensatory, and related to the presence of atherosclerosis. Expansive remodelling in subsegments with higher baseline ESS in the absence of plaque progression likely represents the artery’s physiologic vasodilation response so that shear stress is lowered to a more optimal, vasculoprotective level.

Limitations
Our pilot study is limited by the small number of study subjects. Small numbers and multiple comparisons of data limited statistical power or our ability to detect a statistically significant difference. Therefore, the results of this study are suggestive, but not definitive. The divergent vascular behaviours we observed, however, are consistent with previous in vitro and cross-sectional studies. We are also limited by studying patients at only two points in times, 8 ± 2-months apart, and it is likely that more substantial results would have been observed with a longer follow-up period. We also only studied arteries with minimal lumen obstruction and, consequently, we were not able to investigate the effects of pathologically increased shear stress in more stenosed arteries.

Implications
Plaque progression in minimally obstructed coronary arteries occurs primarily in arterial subsegments with low ESS, and constrictive or expansive remodelling is likely to occur in these same subsegments. These pilot observations will need to be confirmed using a larger sample size. Characterization of the local haemodynamic environment responsible for plaque progression and the local remodelling response may allow for identification of early lesions evolving towards high-risk plaque, providing a rationale for a paradigm shift towards pre-emptive management strategies, such as implantation of drug-eluting stents, to avert adverse cardiac events.

Supplementary material
Supplementary material is available at European Heart Journal online.

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References


