Assessment of atherosclerotic plaques at coronary bifurcations with multidetector computed tomography angiography and intravascular ultrasound-virtual histology

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Received 5 January 2012; accepted after revision 29 March 2012; online publish-ahead-of-print 24 April 2012

Aims
We evaluated the distribution and composition of atherosclerotic plaques at bifurcations with intravascular ultrasound-virtual histology (IVUS-VH) and multidetector computed tomography (MDCT) in relation to the bifurcation angle (BA).

Methods and results
In 33 patients (age 63 ± 11 years, 79% male) imaged with IVUS-VH and MDCT, 33 bifurcations were matched and studied. The analysed main vessel was divided into a 5 mm proximal segment, the in-bifurcation segment, and a 5 mm distal segment. Plaque contours were manually traced on MDCT and IVUS-VH. Plaques with 10% confluent necrotic core and 10% dense calcium on IVUS-VH were considered high risk, whereas plaque composition by MDCT was graded as non-calcified, calcified, or mixed. The maximum BA between the main vessel and the side branch was measured on diastolic MDCT data sets. Overall the mean plaque area decreased from the proximal to the distal segment [8.5 ± 2.8 vs. 6.0 ± 3.0 mm² (P < 0.001) by IVUS-VH and 9.0 ± 2.6 vs. 6.5 ± 2.5 mm² (P < 0.001) by MDCT]. Similarly, the necrotic core area was higher in the proximal compared with the distal segment (1.12 ± 0.7 vs. 0.71 ± 0.7 mm², P = 0.001). The proximal segment had the higher percentage of high-risk plaques (13/25, 52%), followed by the in-bifurcation (6/25, 24%), and the distal segment (6/25, 24%); these plaques were characterized by MDCT as non-calcified (72%) or mixed (28%). The presence of high-risk and non-calcified plaques in the proximal segment was associated with higher BA values (71 ± 19° vs. 55 ± 19°, P = 0.028 and 74 ± 20° vs. 50 ± 14°, P = 0.001, respectively).

Conclusion
The proximal segment of bifurcations is more likely to contain high-risk plaques, especially when the branching angle is wide.

Keywords
Bifurcation • High-risk coronary plaques • Computed tomography coronary angiography • IVUS virtual histology

Introduction
In patients with an atherogenic profile, plaque does not develop evenly across the entire coronary tree, but shows a predilection for sites where the laminar blood flow gets disturbed.1 The low-oscillatory endothelial shear stress has been shown to facilitate atherosclerosis and promote the development of plaques with high-risk features;2,3 these phenomena may explain the increased

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Percutaneous treatment of coronary bifurcation lesions remains a challenging task and has been associated with higher restenosis and stent thrombosis rates.\(^{5–8}\) Better understanding of the underlying pathology, such as the tissue distribution and composition, may allow for safer and more efficient treatment strategies. Intravascular ultrasound–virtual histology (IVUS-VH) is an imaging method that uses the spectral analysis of radiofrequency data to encode four different plaque components, allowing for quantification and characterization of atherosclerotic plaque.\(^{9,10}\)

The complex three-dimensional (3D) geometry of coronary artery bifurcations can affect the local haemodynamic conditions and thereby the plaque distribution and composition. This plausible effect mediated by the bifurcation angle (BA) can only be studied in vivo by means of imaging modalities that provide a 3D reconstruction of the bifurcation, such as multidetector computed tomography (MDCT). MDCT coronary angiography has emerged as a means of non-invasive evaluation of coronary atherosclerotic plaques; its ability to assess the plaque burden, remodelling, eccentricity, and composition has been based on extensive cross-sectional correlation with IVUS data in both stable and unstable patients.\(^{11–13}\)

The objective of the current study is two-fold: (i) to assess in vivo the distribution, composition, and morphology of plaques at bifurcation sites using MDCT and IVUS-VH, and (ii) to explore any possible association of the BA with plaque distribution and composition.

**Methods**

**Study population**

All patients admitted to our hospital between March 2008 and March 2010 who underwent both IVUS-VH and MDCT within a 2-month interval were retrospectively screened for bifurcations adequately visualized by both imaging techniques. The indication for the IVUS-VH was the assessment of angiographically intermediate lesions and/or the result of stent implantation; MDCT was performed for either clinical or research purposes. All patients gave informed consent. Only bifurcations visualized with high quality by both imaging techniques and located at a distance >10 mm from adjacent stents were considered for inclusion.

**Bifurcation selection and matching**

Bifurcation sites involving a side branch (SB) with an ostial diameter ≥1.5 mm on MDCT were only considered for inclusion; left main coronary artery bifurcations were not part of this analysis, because of their entirely different morphology.\(^{14}\) To avoid multiple observations, only a single bifurcation site per patient, the one with the largest SB diameter, was analysed. To ensure proper matching between IVUS-VH and MDCT, the identical bifurcations were identified using the coronary ostia and other bifurcations as landmarks. We chose to analyse only the main branch; the region of interest (ROI) comprised: (i) the proximal segment, extending 5 mm upstream from the proximal take-off of the SB; (ii) the in-bifurcation segment; and (iii) the distal segment, extending 5 mm downstream from the distal take-off of the SB (Figure 1).

**IVUS-VH acquisition and analysis**

The IVUS-VH imaging was performed using the Eagle Eye 20 MHz catheter (Volcano Corp., Rancho Cordova, CA, USA) with an automatic continuous pullback at a rate of 0.5 mm/s. Grayscale images and radiofrequency data required for VH analysis were acquired during the same pullback. The VH data processing was performed offline by an experienced cardiologist with the VIAS software (Volcano Corp., Rancho Cordova, CA, USA) that allows semi-automated contour detection and provides the compositional analysis.

Quantitative IVUS measurements included the vessel area, lumen area, plaque area (vessel area minus lumen area), and plaque burden % [(plaque area/vessel area)×100]. For the radiofrequency-IVUS analyses, four colour-coded tissue components [necrotic core (NC)—red; dense calcium (DC)—white; fibrous (FT)—dark green; and fibrofatty (FF)—light green] were identified with autoregressive classification systems. For every frame, each individual tissue component was quantified as cross-sectional area and percentage (NC + DC + FT + FF = 100%).\(^{9,15}\) Volumetric and compositional parameters obtained per cross-section were averaged for each bifurcation segment.

**MDCT acquisition**

The patients were screened with a first generation dual-source CT scanner (Somatom Definition, Siemens Healthcare, Forchheim, Germany) or a second generation dual-source CT scanner (Somatom Definition Flash, Siemens Healthcare, Forchheim, Germany) which was available after May 2009. Sublingual nitroglycerin was administered prior to the scan (0.4 mg/dose), provided there were no contraindications; no pre-scan beta-blockers were given. An initial non-enhanced ECG-gated scan (120-kV tube voltage, 75-mAs tube current, and 3-mm slice thickness) was performed to calculate calcium-related scores (Agatston, volume, and equivalent mass).\(^{16–18}\) and was followed by a contrast enhanced CT angiography. The CT angiographic parameters were (i) for the first generation dual-source scanner: 32 × 2 × 0.6 mm collimation with the z-flying focal spot for both detectors, gantry rotation time 330 ms, tube voltage 120 kV, current of 320 to 412 mAs, and (ii) for the second generation dual-source scanner: 64 × 2 × 0.6 mm collimation with the z-flying focal spot for both detectors, gantry rotation time 280 ms, tube voltage 120 kV, current of 320 to 412 mAs.
of 100–120 kV, current of 320–370 mAs. A bolus of iodinated contrast material (370 mgI/mL, Ultravist; Schering, Berlin, Germany), which varied between 60 and 100 mL depending on the expected scan time, was injected intravenously (flow rate, 5.5 mL/s) followed by a 40 mL saline chaser at the same injection rate. A bolus tracking technique was used to synchronize the arrival of contrast in the coronary arteries and the start of the MDCT acquisition. For acquisitions with the first generation dual-source CT scanner, retrospective ECG-gated technique with ECG-pulsing was used; when scanning with the second generation dual-source CT either the prospectively ECG-triggered axial scan mode (‘step-and-shoot’) or the retrospective ECG-gated spiral scan mode with ECG-pulsing was used, depending on the heart rate. The pitch for the retrospective scan was set automatically by the scanner software, prior to scanning. The mean effective radiation dose was 9.0 ± 3.9 mSv, using the dose-length product and a conversion factor $k$ of 0.014 mSv/mGy/cm. All MDCT coronary angiograms were reconstructed with a slice thickness 0.75 mm, an increment 0.4 mm, and a medium-to-smooth (B26f) convolution kernel. Optimal data sets with the best image quality were reconstructed mainly in the mid- to end-diastolic phase.

MDCT image analysis

All data sets from the MDCT angiography scans were transferred to a dedicated workstation (Leonardo; Siemens Medical Systems, Erlangen, Germany) for further analysis by an experienced observer, blinded to the results of the IVUS-VH analysis. After the identification of the bifurcation ROI in the original cross-sectional images, serial multiplanar reformatted images (1.0-mm slice thickness, interval 0.5-mm) orthogonal to the longitudinal axis of the main vessel were rendered to obtain cross-sectional images of the respective vessel segment (Figure 2A). The settings for the window level and width were previously optimized by an independent investigator and fixed at 740 and 220 HU, respectively (Rengo M. et al, submitted). Subsequently, the lumen and vessel areas were manually traced in each image, and the plaque areas were calculated as the difference between the vessel and lumen areas (Figure 2D). The plaque areas measured on each cross-section were averaged for each segment and reported as the mean areas.

Plaque type classification

For each one of the three bifurcation segments, the existing plaque phenotype was examined by both modalities. Based on IVUS-VH, lesions were considered present when the percentage plaque area was ≥40% on three consecutive frames. Plaques with >10% confluent NC and <10% dense calcium on three consecutive frames were classified as high-risk (i.e. NC rich) and represented thick-capped fibroatheromas (VH-ThCFAs) or thin-capped fibroatheromas...
which are considered high-risk plaques according to the American Heart Association and Virmani classifications.\textsuperscript{21,22} Based on MDCT, plaques were classified into three types: calcified plaques (≥50% of the plaque area occupied by calcified tissue), mixed plaques (<50% of the plaque area occupied by calcified tissue), and non-calcified plaques which did not contain any calcium at all.\textsuperscript{11}

**Measurement of bifurcation angles with MDCT**

Multiplanar reconstructions (MPRs) were rendered exactly in the plane defined by the main vessel and SB at the bifurcation site, as previously described.\textsuperscript{23} The MPR view where the angulation between the main vessel and SB was maximal was used to determine the BA values. The angle was delineated by two centreline vectors drawn along the initial 5-mm course of the distal main vessel and SB, respectively (Figure 3); only diastolic data sets were used for BA measurements.

**Statistical analysis**

Continuous variables were presented as means ± 1 SD, unless otherwise indicated, and categorical variables were reported as counts and/or percentages. The distribution of data was examined with the Shapiro-Wilk test of normality. Continuous variables were compared between different bifurcation segments and between the two modalities with the paired samples \( t \)-test or the Wilcoxon signed ranks test for two-dependent samples as appropriate. The BA values were compared between groups using the unpaired \( t \)-test. A two-sided \( P \)-value <0.05 was considered statistically significant. Statistical analyses were performed using SPSS 17.0 for Windows (SPSS, Inc., Chicago, IL, USA).

**Results**

**Clinical and procedural characteristics**

Out of 64 consecutive patients with IVUS-VH and MDCT imaging who were screened, 38 were eligible for inclusion. Reasons for exclusion were poor MDCT or IVUS-VH image quality (\( n = 6 \)), presence of stents within the ROI (\( n = 9 \)), and lack of true bifurcations (diameter of the SB ostium <1.5 mm) (\( n = 11 \)). Moreover, in five patients matching of bifurcations between the two imaging modalities was not possible, because more than one bifurcation were located in proximity to each other and could not be reliably identified for the analysis. A total of 33 bifurcation sites from 33 patients were analysed; the mean age was 63 ± 11 years, and 79% of the patients were male. The patients' characteristics are summarized in Table 1. Regarding the bifurcation location, 14 left anterior descending coronary artery/diagonal (42%), 8 left circumflex coronary artery/marginal (24%), 8 right coronary artery/right

![Figure 3](image-url)  
**Figure 3** Coronary bifurcation angle measurements with MDCT. Multiplanar reconstructions were rendered exactly in the plane described by the main vessel and side branch at the bifurcation site. Examples of the bifurcation angles between LAD and D1 (A, 36° and B, 45°) and between RCA and RVB (C, 66° and D, 98°). D1, first diagonal branch; LAD, left anterior descending coronary artery; MDCT, multidetector computed tomography; RCA, right coronary artery; RVB, right ventricular branch.
ventricular branch (24%), and 3 right coronary artery/acute marginal branch (9%) were studied.

Plaque volumetric and compositional characteristics

Overall, the mean plaque area was decreased in the proximal compared with the distal segment (IVUS-VH: 8.5 ± 2.8 vs. 6.0 ± 3.0 mm², \( P < 0.001 \) and MDCT: 9.0 ± 2.6 vs. 6.5 ± 2.5 mm², \( P < 0.001 \), respectively). The mean plaque burden % was higher in the proximal compared with the distal segment (IVUS-VH: 52 ± 13 vs. 44% ± 15%, \( P = 0.002 \) and MDCT 51 ± 11 vs. 46% ± 11%, \( P = 0.029 \), respectively). The volumetric data (Table 2) did not differ significantly between the two modalities. Finally, the mean NC area and the mean percentage of NC were decreased in the proximal compared with the distal segment (Figure 4).

Frequency of plaque type

Based on IVUS-VH, 2 bifurcations (6%) had no atherosclerotic plaques at all, whereas in 14 bifurcations (42%) the plaque was contiguous from the proximal to the distal segment. In total there were 69 bifurcation segments with plaques, where of 25 were high-risk plaques. The proximal segment had the higher percentage of high-risk plaques (13 of 25, 52%), followed by the in-bifurcation segment (6 of 25, 24%) and the distal segment.

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**Table 1** Baseline patients’ characteristics, \( n = 33 \)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>( n = 33 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (mean ± SD), years</td>
<td>63 ± 11</td>
</tr>
<tr>
<td>Male gender, ( n ) (%)</td>
<td>26 (79)</td>
</tr>
<tr>
<td>Risk factors</td>
<td></td>
</tr>
<tr>
<td>Hypertension, ( n ) (%)</td>
<td>24 (73)</td>
</tr>
<tr>
<td>Hypercholesterolaemia, ( n ) (%)</td>
<td>27 (82)</td>
</tr>
<tr>
<td>Diabetes mellitus, ( n ) (%)</td>
<td>6 (18)</td>
</tr>
<tr>
<td>Current smoking, ( n ) (%)</td>
<td>7 (21)</td>
</tr>
<tr>
<td>Family history of CAD, ( n ) (%)</td>
<td>15 (45)</td>
</tr>
<tr>
<td>Previous ACS, ( n ) (%)</td>
<td>10 (30)</td>
</tr>
<tr>
<td>Previous PCI, ( n ) (%)</td>
<td>8 (24)</td>
</tr>
<tr>
<td>Clinical presentation</td>
<td></td>
</tr>
<tr>
<td>Stable angina, ( n ) (%)</td>
<td>24 (73)</td>
</tr>
<tr>
<td>Unstable angina, ( n ) (%)</td>
<td>6 (18)</td>
</tr>
<tr>
<td>Acute myocardial infarction, ( n )</td>
<td>3 (9)</td>
</tr>
<tr>
<td>Studied vessel</td>
<td></td>
</tr>
<tr>
<td>Left anterior descending, ( n ) (%)</td>
<td>14 (42)</td>
</tr>
<tr>
<td>Left circumflex, ( n ) (%)</td>
<td>8 (24)</td>
</tr>
<tr>
<td>Right coronary artery, ( n ) (%)</td>
<td>11 (34)</td>
</tr>
<tr>
<td>Vessel disease, ( n ) (%)</td>
<td></td>
</tr>
<tr>
<td>One vessel disease</td>
<td>16 (50)</td>
</tr>
<tr>
<td>Two vessel disease</td>
<td>13 (41)</td>
</tr>
<tr>
<td>Three vessel disease</td>
<td>3 (9)</td>
</tr>
<tr>
<td>Calcium-related scores*</td>
<td></td>
</tr>
<tr>
<td>Agatston score</td>
<td>313 (147–842)</td>
</tr>
<tr>
<td>Equivalent mass, mg</td>
<td>59 (29–152)</td>
</tr>
<tr>
<td>Volume, mm³</td>
<td>259 (150–701)</td>
</tr>
</tbody>
</table>

*Values are median (inter-quartile range). ACS, acute coronary syndrome; CAD, coronary artery disease; PCI, percutaneous coronary intervention; SD, standard deviation.

**Table 2** Volumetric data in each segment of the bifurcation by MDCT and IVUS-VH

<table>
<thead>
<tr>
<th>Parameter</th>
<th>MDCT (mean ± SD)</th>
<th>IVUS-VH (mean ± SD)</th>
<th>( P )-value (t-test)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean plaque area (mm²)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proximal</td>
<td>9.0 ± 2.6</td>
<td>8.5 ± 2.8</td>
<td>0.09</td>
</tr>
<tr>
<td>In-bifurcation</td>
<td>7.2 ± 2.4</td>
<td>7.4 ± 3.0</td>
<td>0.94</td>
</tr>
<tr>
<td>Distal</td>
<td>6.5 ± 2.5</td>
<td>6.0 ± 3.0</td>
<td>0.09</td>
</tr>
<tr>
<td>Mean plaque burden (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Proximal</td>
<td>51 ± 11</td>
<td>52 ± 13</td>
<td>0.69</td>
</tr>
<tr>
<td>In-bifurcation</td>
<td>46 ± 11</td>
<td>45 ± 13</td>
<td>0.20</td>
</tr>
<tr>
<td>Distal</td>
<td>46 ± 11</td>
<td>44 ± 15</td>
<td>0.15</td>
</tr>
</tbody>
</table>

IVUS-VH, intravascular ultrasound-virtual histology; MDCT, multidetector computed tomography; SD, standard deviation.

**Figure 4** Necrotic core (NC) distribution. NC area (A) and NC percentage (B) distribution in the proximal, in-bifurcation, and distal segments.
(6 of 25, 24%); the distribution of the different plaque types within each segment is shown in Figure 5A. On MDCT, non-calcified plaques presented more frequently in the proximal segment, whereas the calcified plaques were more frequent in the distal segment (Figure 5B). The majority of high-risk plaques as determined by IVUS-VH were characterized by MDCT as non-calcified ones (72%); the remaining 28% were classified as mixed. The plaque type distribution per segment is shown in Figure 6.

### Plaque distribution and composition in relation to the bifurcation angle

Bifurcations containing plaques in their proximal segments had significantly larger BA values compared with those without \((P = 0.002)\), Table 3. Moreover, the presence of high-risk and non-calcified plaques in the proximal segment was associated with higher BA values \((71 \pm 19 \text{ vs. } 55 \pm 19, P = 0.028 \text{ and } 74 \pm 20 \text{ vs. } 50 \pm 14, P = 0.001, \text{ respectively})\). The BA was not related significantly with either plaque distribution or composition in the other segments.

### Discussion

In this exploratory study, we evaluated the distribution and composition of atherosclerotic plaques at coronary bifurcations using invasive and non-invasive imaging. To our knowledge, this is the first in vivo study to examine the volumetric and compositional plaque characteristics in combination with the 3D geometry of coronary bifurcations (as expressed by the BA).

Our main findings can be summarized as follows: (i) the proximal segment has a more extensive plaque burden with more NC; (ii) the plaques with high-risk features on IVUS-VH and the non-calcified plaques on MDCT both show a differential distribution along the bifurcation being more frequent in the proximal segment; (iii) the distribution and composition of plaques in the proximal segment is associated with the BA values.
The current study corroborates earlier findings on plaque volumetric and compositional characteristics at bifurcation sites. In a previous IVUS study, Badak et al.\textsuperscript{24} suggested that significantly more atheroma was located proximal to the bifurcation than distally. Han et al.\textsuperscript{19} expanded on these findings using IVUS-VH to report on tissue characterization; that study showed that the plaque burden and the percentage of the NC were significantly larger in the proximal segment of the non-left main bifurcations. These data appear to be largely concordant with a prior report by Gonzalo et al.,\textsuperscript{5} using IVUS-VH and Optical Coherence Tomography (OCT); they suggested that the percentage of the NC was higher at the proximal ‘rim’ of the bifurcation. It has been shown that the distribution of inflammatory cells in atherosclerotic plaques is associated with the direction of the arterial flow, with higher content of macrophages in the upstream (proximal) part.\textsuperscript{25} The differential distribution of the NC and the high-risk plaques along the coronary tree could be attributed to the influence of local haemodynamic factors altered at bifurcation sites.

Our data demonstrated that predominantly non-calcified plaque type by MDCT corresponded to the IVUS-VH high-risk phenotype. This finding is in agreement with histopathology studies\textsuperscript{22} and with previous greyscale IVUS studies showing that plaques with low echogenicity (presumably lipid rich) are mainly soft (non-calcified) by MDCT.\textsuperscript{11,26,27} Conversely, other studies suggested that mixed plaques on MDCT are associated more frequently with high-risk features on IVUS-VH.\textsuperscript{28,29} A recent more systematic evaluation\textsuperscript{30} demonstrated that only the non-calcified plaques with small (<1 mm) spotty calcifications on MDCT were associated with plaque characteristics deemed more high risk on IVUS-VH; however, these plaques did not have significantly more VH-TCFAs compared with non-calcified plaques. In our analysis, the ROI included only plaques located within 5-mm long segments proximal and distal to the SB; since atherosclerotic plaque can be diffuse, in case a spotty calcification was located outside this ROI, it would have not been included in our analysis.

The present study extended the assessment of bifurcation lesions beyond volumetric and compositional analysis by integrating the MDCT-based BA measurements, which emphasizes the added value of this 3D imaging modality. Our in vivo data showed that the presence and phenotype of atherosclerotic plaque in the proximal bifurcation segment is related to the BA size. This finding could be supported by computational fluid dynamics\textsuperscript{31–33} and histopathology\textsuperscript{34} studies demonstrating that the haemodynamic phenomena important in atherogenesis are more pronounced in widely angulated bifurcations. Moreover, the BA size has been described as a determinant of treatment strategy in bifurcation lesions.\textsuperscript{35} In numerous bench and clinical studies,\textsuperscript{36–40} a wide BA has been associated with a greater risk for suboptimal post-procedural result and long-term adverse clinical events. Additionally, our data demonstrated that a wide BA is associated with a greater plaque burden and a high-risk phenotype of bifurcation lesions, which make us speculate that NC-rich plaques could be related to the higher restenosis and thrombosis rates.\textsuperscript{41–43} Eventually, a comprehensive assessment of bifurcation lesions including plaque characterization and BA measurements could lead to optimized interventional strategies and improved long-term clinical outcomes.

**Limitations**

Although this is the first study combining invasive and non-invasive imaging of bifurcations, it was retrospectively performed in a selected patient population undergoing invasive and non-invasive imaging, thus generalization of our results should be done with caution. Given the inclusion of a limited number of patients, the possibility of a type II error should be considered and multivariate analysis for risk factors was not performed. Furthermore, the limited axial resolution of the IVUS (≏200 μm) does not permit an accurate evaluation of the thin fibrous cap (<65 μm). A proposed method of combined use of IVUS-VH and OCT\textsuperscript{3,44} could facilitate more accurate identification of VH-TCFAs, but was not available for this study. Finally, the radiation exposure during MDCT coronary angiography remains a matter of concern; significant reduction in radiation dose can currently be achieved by implementation of dose-saving techniques.\textsuperscript{45}

**Conclusions**

This study extended the evaluation of bifurcation lesions beyond volumetric and compositional analysis by integrating information on the geometry of coronary bifurcations. A larger plaque burden and increased NC content were found in the proximal bifurcation segments. The high-risk plaques on IVUS-VH and the non-calcified plaques on MDCT tend to accumulate in the proximal bifurcation segments, especially in the presence of a wide BA.

**Conflict of interest:** None declared.

**References**


