Angioscopic differences in neointimal coverage and in persistence of thrombus between sirolimus-eluting stents and bare metal stents after a 6-month implantation

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Aims The neointimal coverage and intracoronary thrombi within stented segments at 6 months after implantation between sirolimus-eluting stents (SESs) and bare metal stents (BMSs) were compared by direct visualization using angioscopy.

Methods and results Forty-six patients (36 stable angina and 10 acute coronary syndrome) were treated with 33 SESs and 33 BMSs. Immediately after and 6 months after stenting, each of the stented segments, edge body, and overlapping segment were observed by angioscopy and the grade of neointimal coverage over the stents was classified as 0: absent neointima, 1: visible struts through thin neointima, or 2: invisible struts. The existence of thrombi was also evaluated. The average grade of the neointimal coverage at 6 months follow-up was lower in the SES than that in the BMS (edge: 1.4 ± 0.7 vs. 1.9 ± 0.2, body: 1.0 ± 0.5 vs. 1.8 ± 0.5, overlapping segment: 0.6 ± 0.7 vs. 1.8 ± 0.5; P < 0.0001, P < 0.0001, P = 0.0069, respectively). The frequency of persistence of thrombus was significantly higher in the SESs than that in the BMSs (86 vs. 29%, respectively; P = 0.031).

Conclusion The present study suggested a delayed neointimal stent coverage and slower thrombus disappearance process in the SESs in comparison to the BMSs.

KEYWORDS
Stent;
Neointimal hyperplasia;
Thrombus

Introduction
To date, stent implantation has become the standard therapy in percutaneous coronary intervention (PCI) for patients with atherosclerotic coronary disease. Nevertheless, in-stent restenosis (ISR) within 3–8 months is a factor that limits the long-term success of stenting and it is mainly caused by neointimal hyperplasia.1 Recently, sirolimus, a cytostatic macrocyclic lactone with both anti-inflammatory and antiproliferative properties, delivered from a polymer-encapsulated stent was shown in several angiographic and intravascular ultrasound (IVUS) studies to reduce the risk of ISR in comparison with the bare metal stent (BMS).2–8 Neointimal proliferation inside the sirolimus-eluting stent (SES), which is recognized as lumen late loss on angiograms or as an obstruction volume on the IVUS, was minimal and nearly abolished at the 6-month follow-up.4–7 Moreover, the in-stent lumen dimensions remained essentially unchanged for a long-term without a ‘late catch-up’.9 In contrast, there have been recent concerns regarding their potential for developing late-stent thrombosis related to the delayed re-endothelialization over the struts of the SES, particularly after discontinuation of dual antiplatelet therapy.10,11 Therefore, it is important to know when complete re-endothelialization of drug-eluting stent occurs. If complete neointimal coverage of the drug-eluting stent is accomplished, thienopyridine may no longer be needed. Coronary angioscopy provides a direct visualization of the luminal surface and detailed information on the state of stent coverage by the neointima, changes in the plaque colour, and the existence of intracoronary thrombi.11–15 In this study, the condition of neointimal coverage and intracoronary thrombi over SESs are compared with those observed in BMSs at 6 months after their implantation.

Methods
Patients population
Between October 2004 and April 2005, 53 patients with de novo and native coronary artery lesions were treated with SESs (Cypher, Cordis Corp., Miami Lakes, FL, USA). During this study period,
neither glycoprotein IIb/IIIa inhibitors nor clopidogrel had been approved for clinical use in Japan. Beforehand, all patients received two kinds of antiplatelet agents, ticlopidine 200 mg/day (standard dose under clinical approval in Japan) and aspirin 100-200 mg/day, longer than 48 h before the PCI for the prevention of acute or subacute thrombosis. Exclusion criteria were (i) acute myocardial infarction within 48 h from onset \( n = 3 \), (ii) restenotic lesions after balloon angioplasty or ISR \( n = 7 \), (iii) a low ejection fraction of the left ventricle \( n = 2 \), (iv) left main disease or ostial lesions \( n = 12 \), and (v) tortuous vessels or heavily calcified vessels proximal to the culprit lesions \( n = 8 \) because of the expected difficulty in acquiring angioscopic images for the whole stented segments or in advancing the angioscopic catheter. Finally, 21 patients were enrolled in the SES group. Between October 2000 and May 2001, 61 patients were treated with BMSs. According to the above exclusion criterion, 36 patients (20 acute myocardial infarction, five restenosis, three low ejection fraction, four ostial lesions, and four vessel tortuosity or calcification) were excluded. Consequently, 25 patients were selected as the BMS group. All patients included this study received follow-up angiography and angioscopy at 6 months. Written informed consent approved by our institutional review board was obtained from all study patients before catheterization.

Clinical demographics

The patient demographics were obtained by a hospital chart review. Stable angina pectoris (SAP) was defined as a positive stress test and no change in the frequency, duration, or intensity of symptoms within 4 weeks. Unstable angina was new-onset severe angina, accelerated angina, or rest angina. Recent myocardial infarction was defined as its occurrence between 2 days and 2 weeks before the PCI. Patients with acute myocardial infarction less than 2 days from onset were not enrolled. Patients with unstable angina and recent myocardial infarction were categorized as acute coronary syndrome (ACS).

Culprit lesions (target lesions of the PCI) were identified by a combination of the ECG findings, wall motion abnormalities on left ventriculography or the echocardiography findings, and angiographic lesion morphology.

Angiographic analysis

All angiograms were analysed with a computer-assisted, automated edge detection algorithm (CMS, MEDIS, Nuenen, The Netherlands) by an angiographer blinded to the clinical and angioscopic findings using standard qualitative definition and quantitative coronary angiogram measurements. A 6-month follow-up angiogram was performed at the same angle as the PCI. The reference vessel diameter (RVD), minimal lumen diameter (MLD), percent diameter stenosis (%DS), lesion length, and late loss at the culprit lesion or stented segment were measured. Angiographic filling defects, haziness, or wall irregularity was qualitatively evaluated. ISR was defined as more than or equal to 50% of DS at follow-up, located within the stented segments and in the segments adjacent to the proximal and distal edge of the stent.

Coronary angioscopic imaging

The coronary angioscopic procedure has been previously reported. Angiographic examinations were performed before PCI, immediately after PCI, and 6 months after PCI as follow-up studies. Culprit plaques and whole-stented segments were observed with an angioscopic catheter (Vecmova Neo, Clinical Supply Corp., Gifu, Japan). The angioscopic images and fluoroscopy during the angioscopic observations were recorded on digital videotape for later analysis. The exact position of the angioscopic catheter at the observed segment was recorded by an angiogram to ensure a reliable comparison.

Clinical follow-up

Two kinds of antiplatelet agents, ticlopidine (200 mg/day) added to aspirin (100-200 mg/day), were administered continuously during the 6-month follow-up period. Repeat PCI, coronary bypass surgery, the occurrence of ACS, and cardiac sudden death were all considered to be major adverse events.

Statistical analysis

The statistical analysis was performed using StatView 5.0 (SAS Institute). Categorical variables are presented as frequencies and
they were analysed by either the $\chi^2$ test or the Fisher’s exact test. Continuous quantitative data and discontinuous data (angioscop ic grades for plaque colour and stent coverage) are presented as mean ± SD. Continuous data were compared by the unpaired Student’s $t$-test between the different categories. Ordinal data of stent coverage grade were tested by the Mann-Whitney U test with the Bonferroni’s correction between the different categories (between different segments in the same kind of stent and corresponding segment between different kinds of stents). The yellow grade among the same categories between those observed at baseline and at follow-up was compared by the Wilcoxon singed-rank test or the Fisher’s exact test.

### Results

#### Clinical characteristics at baseline

Thirty-three SESs were implanted in 21 lesions of 21 patients and 33 BMSs were implanted in 28 lesions of 25 patients. The patients’ characteristics in the SES group ($n = 21$) and those in the BMS group ($n = 25$) at baseline are summarized in Table 1. No significant difference in the age, proportion of gender, risk factors of atherosclerosis, including diabetes mellitus, diagnosis for coronary artery disease (SAP or ACS), or medications, existed between the two groups.

#### Angiographic findings

The angiographic data are summarized in Table 2. The lesion location did not differ between the two groups. Immediately after PCI, both of MLD and acute gain were greater in the BMS group than that in the SES group. At the 6-month follow-up, both the %DS and late loss were smaller in the SES group than that in the BMS group. There were no filling defects, haziness, or wall irregularity in neither of the two groups at follow-up. Despite the fact that one case in the SES group showed focal ISR, no edge restenosis was seen on the angiogram. The SES group tended to show a decreased RVD in comparison to the SES group (5 vs. 0.5%, $P = 0.07$).

### Angioscopic findings

Angioscopic findings at baseline (before and immediately after PCI) and at the 6-month follow-up are summarized in Table 3. Before PCI, the frequency of plaque rupture and yellow grade of the culprit plaque did not differ between the two groups. At baseline, thrombus was observed in seven patients and in seven BMS patients (five in ACS and two in SAP), and the frequency of the thrombus was similar between the two groups (33 vs. 28%, respectively). At the 6-month follow-up, the yellow grade of the culprit plaque was higher in the SES group than that in the BMS group (1.1 ± 0.5 vs. 0.5 ± 0.6, $P = 0.0007$). The relationship between the yellow grade regression and the frequency of the patients is presented in Figure 2.
The yellow grade regression of the culprit plaque was lower in the SES group than that in the BMS group (0.3 ± 0.6 vs. 1.1 ± 0.7, \( P < 0.0001 \)). In the SES group, six of seven thrombi found at baseline remained at the 6-month follow-up, and one thrombus was newly recognized at follow-up. As a result, seven thrombi in the SES group were observed at follow-up. In the BMS group, two of seven thrombi found at baseline remained at the 6-month follow-up, and there was no thrombus formation during the 6-month follow-up period. The frequency of persistence of thrombus was higher in the SES group than that in the BMS group (86 vs. 29%, respectively; \( P = 0.031 \)) (Figure 3). The angioscopic and angiographic findings for each case in the SES and the BMS groups are shown in Figure 4.

In the SES group, a single SES was deployed in 11 lesions, two SESs were placed in eight lesions, and three SESs were used in two lesions. In the BMS group, a single BMS was deployed in 23 lesions and two BMSs were placed in five lesions. In both of the groups, multiple stent deployment for a single lesion completely overlapped without a gap. At the 6-month follow-up, 21 stent edges, 33 stent bodies, and the 12 stent overlapping segments of the SES group were analysed for the stent coverage grade. Similarly, 28 stent edges, 33 stent bodies, and five stent overlapping segments of the BMS group were analysed. In all segments, the stent coverage grade for the SES was lower than that for the BMS (1.1 ± 0.7 vs. 1.9 ± 0.4, respectively; \( P < 0.0001 \)).

### Table 3 Angioscopic findings at baseline and follow-up

<table>
<thead>
<tr>
<th>Patients</th>
<th>SES (n = 21)</th>
<th>BMS (n = 25)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Plaque rupture (Before PCI)</td>
<td>4 (19%)</td>
<td>5 (20%)</td>
<td>0.94</td>
</tr>
<tr>
<td>Yellow grade of the culprit plaque</td>
<td>1.4 ± 0.6</td>
<td>1.6 ± 0.9</td>
<td>0.39</td>
</tr>
<tr>
<td>Thrombus</td>
<td>7 (33%)</td>
<td>7 (28%)</td>
<td>0.022</td>
</tr>
<tr>
<td>Six-month follow-up</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yellow grade of the culprit plaque</td>
<td>1.1 ± 0.5</td>
<td>0.5 ± 0.6</td>
<td>0.0007</td>
</tr>
<tr>
<td>Thrombus</td>
<td>7 (33%)</td>
<td>2 (8%)</td>
<td>0.049</td>
</tr>
</tbody>
</table>

Values are n (%) or mean ± SD.

![Figure 2](https://example.com/figure2.png)  
**Figure 2** The relationship between the yellow grade regression of the culprit plaque and the frequency of the patients. In the SES group, the yellow grade regression of –1, 0, 1, and 2 were accounted for 5, 52, 43, and 0% of the patients, respectively. In the BMS group, that of –1, 0, 1, and 2 were accounted for 0, 12, 52, and 36% of the patients, respectively.

![Figure 4](https://example.com/figure4.png)  
**Figure 4** Two cases showing the angiographic and angioscopic findings after stent implantation in thrombotic lesions. (A) Immediately after SES implantation in the left circumflex artery in a patient with ACS. A pinkish mural thrombus was clearly found (between 0 and 5 o’clock) beside the plaque of the yellow grade 3 (at 11 o’clock). (B) A follow-up exam at 6 months in the case shown in (A). ISR was not seen on the angiogram. Residual thrombus was recognized (between 0 and 2 o’clock). Parts of the struts were absent in the neointimal coverage (the stent coverage grade 0), and a thin neointimal proliferation on the plaque of the yellow grade 2 (between 2 and 4 o’clock) was found. The yellow grade regression was 1 in this case. (C) Immediately after BMS implantation in the left anterior descending artery in a patient with ACS. A pinkish mural thrombus was found (between 0 and 8 o’clock) beside the plaque of the yellow grade 2 (between 0 and 3 o’clock). (D) A follow-up exam at 6 months in the case shown in (C). ISR was not seen on the angiogram. There was no residual thrombus. The struts were completely covered by neointima (the stent coverage grade 2 and the yellow grade 0). The yellow grade regression was 2 in this case.

![Figure 3](https://example.com/figure3.png)  
**Figure 3** A comparison of the frequency of persistence of thrombus between the SES and BMS. The frequency of persistence of thrombus was significantly higher in the SES than in the BMS (86 vs. 29%, respectively).
The stent coverage grade in the SES was 1.4 ± 0.7 in edge, 1.0 ± 0.5 in body, and 0.6 ± 0.7 in overlapping segment. This grade was lower in the body than in the edge (P = 0.0002) and in the overlapping segment than in the body (P = 0.016). In the BMS group, the stent coverage grade in the edge body and overlapping segment was 1.9 ± 0.2, 1.8 ± 0.5, and 1.8 ± 0.5, respectively. No segment was significantly different from another. The stent coverage grade of the SES group was lower than that of the BMS group, in the edge (P < 0.0001), the body (P < 0.0001), and the overlapping segment (P = 0.0069) (Figure 5). A total of 27 segments (41% of all segments; six edges, 15 bodies, and six overlapping segments) of the SES and four segments (6% of all segments; four bodies) of the BMS were evaluated to have an absence of neointimal coverage (grade 0).

Clinical events
All patients underwent successful PCI for culprit lesions at baseline. Four lesions in three patients (12%) in the BMS group and none of the patients in the SES group underwent repeat PCI for ISR 6 months after the first PCI. All patients in both groups were free from bypass surgery, ACS, and cardiac sudden death during the clinical follow-up period.

Discussion
Although several follow-up studies after SES implantation revealed the effect of neointimal proliferation on its inhibition, there is a lack of detailed data regarding changes in underlying atherosclerotic yellow plaque due to neointimal stent coverage and intracoronary thrombus. Coronary angiography provides detailed information on plaque colour, neointimal hyperplasia, and mural thrombus by the direct visualization of the coronary lumen. Our angiographic follow-up study demonstrated that the processes of changes in yellow plaque, neointimal coverage, and thrombus disappearance within the stented segments in the SES occurred more slowly than those in the BMS.

Differences in neointimal coverage between SES and BMS

The grade of neointimal coverage after BMS implantation was previously classified as either 0: complete exposure of stent struts, 1: exposure of the struts with partial coverage, 2: more than 50% coverage, 3: almost complete coverage with slightly visible the struts, or 4: complete coverage. However, it was difficult to determine this grade from 1 to 3 after SES implantation, because neointimal growth over the SES was frequently too thin and was partially transparent. The transparent neointima diagnosed by angioscopy may be layer of fibrin deposition. Therefore, simple grading system of the neointimal coverage was used in this study.

Experimental models of rabbit’s iliac arteries confirmed that SES shows a decrease in neointimal thickness of 26.3% in comparison to BMS (Bx velocity stents), platform of the SES. At the 6-month follow-up, the late loss on the angiogram in the SES was smaller than that in the BMS. The stent coverage grade on the angioscopic findings in the overall stented segments in the SES was lower than that in the BMS. These results thus supported the notion that the neointimal proliferation inside the SES at the chronic phase was inhibited to be thinner than that in the BMS, and this finding was similar to the findings of previous IVUS investigations in living patients.

One of the major factors that influence angioscopic yellow grade is thickness of the fibrous cap or neointima over atherosclerotic lipid contents. Thick neointimal hyperplasia after stenting might change the plaque colour from yellow to white or reduce the yellow grade. Therefore, the differences in the yellow grade regression of the culprit plaque between the SES and the BMS may be explained by the differences in the thickness of the neointimal hyperplasia. The thin neointima over the SES struts on the yellow plaque may make it easier to find the struts through the neointima and it may also be less influential in changing the yellow grade of the plaque.

Differences in neointimal coverage between the different segments

Angiographic neointimal coverage over the BMS at the 6-month follow-up was almost complete and equal on the edge, in the body, and in the overlapping segment of the stents. In contrast, the degree of neointimal coverage of the SES differed between the edge, the body, and the overlapping segment. The neointimal coverage grade of the SES was significantly lower in the overlapping segment than in the body. These angiographic results agree with the findings of a recent pathohistological report by Finn et al. in which they demonstrated that both incomplete endothelialization (delayed endothelialization) as assessed by light microscopy and chronic inflammation were found more commonly in the overlapping segments than in the non-overlapping segments of the SES. They speculated that the poor endothelialization and persistent inflammation in the overlapping segment are based on the contribution of drug overdose, a hypersensitive reaction to the polymer, and/or metal overload. In the present study, 27 of 66 segments (41%) of the SES were evaluated to have an absence of neointimal coverage and an exposure of the struts on angioscopy. A macroscopic diagnosis using...
angioscopy has limitation to detect infiltration of inflammatory cells, tiny fibrin deposition, and very thin neointimal proliferation on the struts.\textsuperscript{17} Other imaging modalities with a high-resolution, such as optical coherence tomography, may help to clarify the fine structure inside the SES.\textsuperscript{17}

Previous IVUS studies revealed that the SES does not create any of the edge effects predominantly caused by negative remodelling with exaggerated neointimal hyperplasia.\textsuperscript{5,7,8} In the present study, angioscopic neointimal coverage grade of the SES was higher in the edge than in the body at follow-up. The precise mechanism of the differences in the degree of neointimal proliferation is not clear. Several speculative hypotheses may be raised to explain this phenomenon. First, at the proximal or distal edges, the drug dose and/or metal dose of the SES is theoretically half of that in the body because the existence of the struts is only on one side, namely either the distal or the proximal side. Secondly, the edges of the SES are usually located in non-stenotic healthy segments, whereas its bodies are in the stenotic diseased segments on the angiogram. Therefore, the composition of the vessel wall (plaque) might differ between the two locations. These differences in the composition of vessel wall (plaque) may be the cause of the differences in the degree of neointimal proliferation on angioscopy.

Differences in thrombus disappearance between SES and BMS

After stent implantation, it has been understood that smooth muscle cells in the media proliferate and migrate to the lumen surface under the influence of growth factors.\textsuperscript{18} However, the processes of thrombus disappearance after stenting in living patients are necessarily less detailed. The neointimal growth involved in the thrombus change to fewer cellular elements and a richer extracellular matrix probably occurs over a period of weeks or months.

In our series, the SES showed more unfavourable effects on the thrombus disappearance within the stented segments than the BMS. Several possible explanations can be offered for these results. First of all, sirolimus directly inhibits the proliferation of smooth muscle cells. As a result, thrombus disappearance may be indirectly delayed due to a suppression of neointimal growth. Secondly, the \textit{in vitro} findings indicated that sirolimus (rapamycin) by itself increases basal tissue factor levels by 40\% in human aortic endothelial cells.\textsuperscript{19} The tissue factor, contained in human atherosclerotic plaques, is well known to be one of the thrombogenic agents. The tissue factor expressed on the endothelium may therefore disturb the thrombus disappearance.

Although angioscopy has limitations to confirm the existence of very thin neointima, macroscopic examinations at the 6-month follow-up failed to reveal a complete neointimal coverage overlying the struts, especially in the overlapping segment of the SES. Moreover, most of the intracoronary thrombi within the SES remained at follow-up. It may thus be necessary to investigate the duration of aggressive antplatelet therapy after SES implantation. In the BMS, the phenomenon that the neointima becomes thick and non-transparent until 6 months after stent implantation and then thin and transparent by 3 years has been previously reported.\textsuperscript{20} Further long-term follow-up studies to elucidate changes in the neointima and the residual thrombi are thus required.

Study limitations

Our findings were based on observations in a relatively small number of patients and stented segments excluding acute myocardial infarction within 48 h from onset. Some selection bias is therefore inevitable. This study was not a randomized study. Therefore, the RVD before the PCI was larger in the BMS group than in the SES group. The patient characteristics between the two groups were well matched. However, under these conditions, the antiproliferative effects of the SES were demonstrated by angioscopy.

Conclusions

This study failed to prove complete neointimal coverage of SES 6 months after implantation, despite macroscopic diagnosis by angioscopy. Our angioscopic study suggested both a delayed neointimal proliferation and a slow process in thrombus disappearance in the SES in comparison with those in the BMS at 6 months after their implantation. Further long-term follow-up studies to clarify the serial changes of the neointima and thrombi inside the SES are thus called for.

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Conflict of interest: none declared.

References

Huge right coronary artery aneurysm in a young adult

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A 34-year-old man visited our institution for an abnormal chest radiograph with greatly enlarged cardiac silhouette (Panel A) found before his wedding. Detailed medical history revealed a febrile episode and a strawberry-like tongue and some ‘flu-like symptoms’ for >1 week when he was 5 years old. Diagnostic workup including computerized tomography demonstrated a huge right coronary artery aneurysm (Panel B). Intra-operative transesophageal echocardiography showed a large orifice of right coronary artery with a huge aneurysm (Panel C). The right coronary artery aneurysm was resected (Panel D) under hypothermic cardiopulmonary bypass with cardioplegic arrest. The diameter of the orifice of the right coronary artery was >20 mm, which was closed with Dacron patch, and the saphenous vein was used to bypass the right coronary artery to its two major branches. The patient had an uneventful recovery and was discharged 6 days after the operation.

Panel A. Chest X-ray shows an enlarged cardiac silhouette.

Panel B. Thorax CT-scan shows the right coronary artery aneurysm (white arrows) and its connection (black arrow) with the aorta. MPA, main pulmonary artery.

Panel C. Transesophageal echocardiography shows the enlarged orifice of right coronary artery (small arrow) and the large aneurysm (large arrow).

Panel D. Surgical views of the huge right coronary artery aneurysm (small arrows) and aorta (large arrow).