

## Diverse Findings in Calcified Thrombus Between Histopathology and *In Vivo* Imaging Including Intravascular Ultrasound, Optical Coherence Tomography, and Angioscopy

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### SUMMARY

A 46-year-old woman on hemodialysis due to end-stage renal disease was admitted for repeated thrombus formation in previously implanted drug-eluting stents in the right coronary artery. We could successfully aspirate this thrombus, and histopathology revealed a calcified thrombus comprising multiple microcalcifications and fibrinous materials. This is the first report showing how a calcified thrombus is visualized *in vivo* by intracoronary imaging modalities including intravascular ultrasound, optical coherence tomography, and angioscopy. (Int Heart J 2015; 56: 661-663)

**Key words:** Stent thrombosis, Intravascular imaging, Coronary artery disease, Drug-eluting stent

Among intracoronary imaging modalities including intravascular ultrasound (IVUS), optical coherence tomography (OCT), and angioscopy, OCT and angioscopy are the most reliable to detect a coronary thrombus and to discriminate red and white thrombus *in vivo*.<sup>1-3)</sup> Here, we present a rare case of in-stent calcified thrombus presenting diverse findings by IVUS, OCT, angioscopy, and histopathology.

### CASE REPORT

A 46-year-old woman on hemodialysis due to end-stage renal disease was admitted with repeated stent thrombosis (ST). She had a history of hypertension, dyslipidemia, type 1 diabetes mellitus, and cigarette smoking. She had initially been treated 3 years previously with the implantation of 3 paclitaxel-eluting stents (PES) for chronic total occlusion of the right coronary artery (Figure 1A - C), along with bypass surgery of the left internal thoracic artery to the left anterior descending artery and of the saphenous vein to the left circumflex artery. She was administered aspirin and clopidogrel thereafter. Two years after PES implantation, a first ST occurred at the distal PES site, where an everolimus-eluting stent (EES) was implanted (Figure 1D). Four months later, a second ST occurred at the EES site within the PES, which was treated by balloon angioplasty. Three months later, a third ST occurred at the same site as the second ST, which was treated with a zotarolimus-eluting stent (ZES) implantation in the EES (Figure 1E). Follow-up angiography 20 days later revealed a filling defect (Figure 1, white arrow) indicating a thrombus formation at the ZES site inside the EES. She was referred to our hospital for

further evaluation and treatment.

She had been taking dual antiplatelet therapy including aspirin and clopidogrel. The platelet function tests were performed using the VerifyNow system (Accumetrics, San Diego, CA, USA). Measures of the antiplatelet effect of aspirin are expressed as aspirin reaction units (ARU), and those of clopidogrel are expressed as P2Y12 reaction units (PRU). Her results of the VerifyNow test were 465 ARU and 153 PRU. Prior to the percutaneous coronary intervention, the IVUS, OCT, and angioscopy were performed for this angiographic filling defect lesion in stent. The IVUS (Atlantis SR Pro 2, Boston Scientific, Natick, MA, USA) showed a hypoechoic mass with a superficial hyperechoic signal (Figure 2A). The OCT (C7 OCT Imaging System, St Jude Medical, St Paul, MN, USA) showed a mass protruding into the lumen with high OCT-signal backscattering and attenuation (Figure 2B). Angioscopy (Fullview NEO, iHeart Medical Co.Ltd., Tokyo) to this lesion revealed a white material with irregular surface protrusion into the lumen (Figure 2C). IVUS, OCT, and angioscopy revealed that most of the stent struts were not covered by apparent neointima (Figures 2D, E, F). Finally, this mass was easily and successfully removed using an aspiration catheter. IVUS, OCT, and angioscopy after aspiration confirmed that the mass was decreased dramatically and enough lumen area was obtained (Figures 2G, H, I) so subsequent procedures such as ballooning and stent implantation were not required. Histopathology of the aspirated mass revealed a calcified thrombus comprising multiple microcalcifications and fibrinous materials without apparent platelets or other blood corpuscles (Figure 3).

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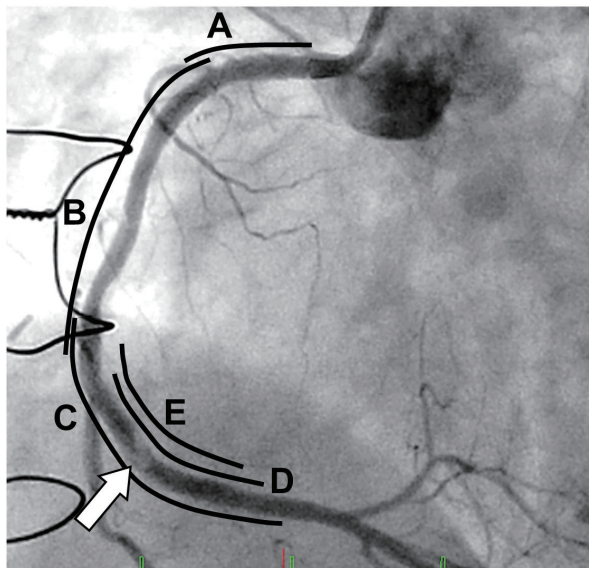
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## DISCUSSION

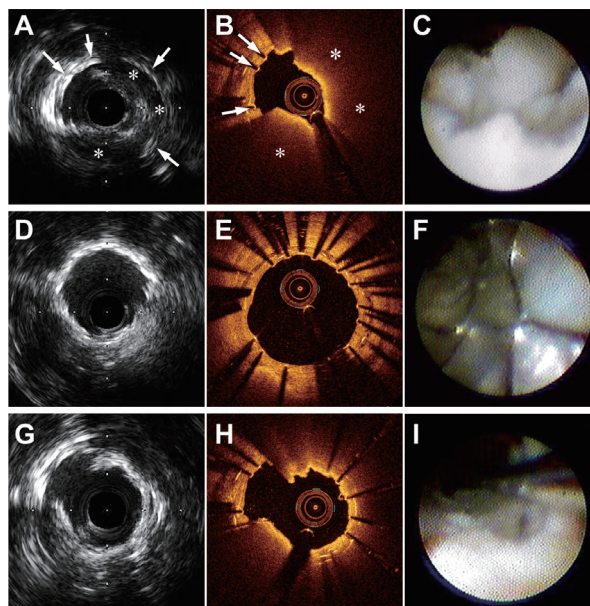
In the present case, a mass aspirated from an angiographic filling defect lesion in stent was diagnosed histopathologically as a calcified thrombus comprising multiple microcalcifications and fibrinous materials. Of note, *in vivo* images of the calcified thrombus were not typical for calcification by either OCT or angioscopy. To the best of our knowledge, this is the first report demonstrating how a calcified thrombus appears *in vivo*.

In this case, IVUS images of the calcified thrombus showed a hypoechoic mass with a superficial hyperechoic signal. This finding may imply the presence of slight calcification.

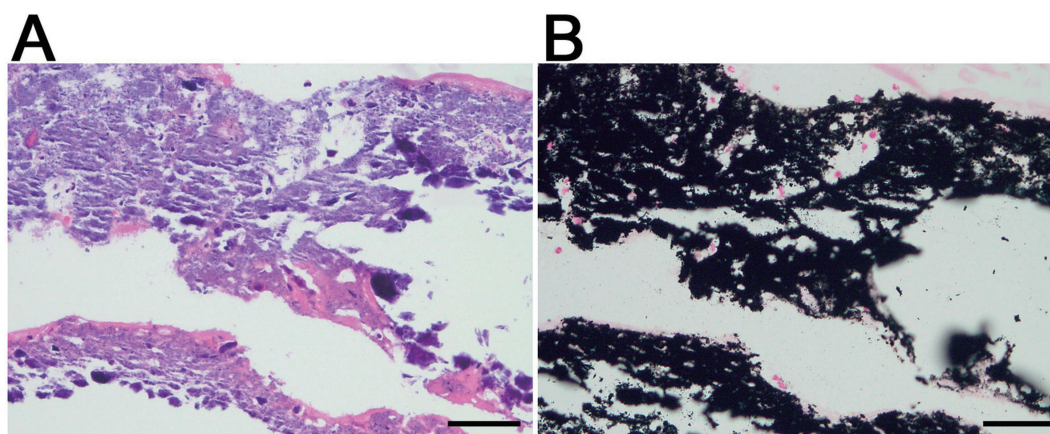


**Figure 1.** Angiographic images of previously implanted stents (black lines) in right coronary artery. **A:** Paclitaxel-eluting stent (PES) 3.5 × 16 mm; **B:** PES 3.0 × 38 mm; **C:** PES 2.75 × 38 mm; **D:** Everolimus-eluting stent (EES) 2.75 × 33 mm in PES; **E:** Zotarolimus-eluting stent 2.75 × 30 mm in PES. A white arrow indicates a filling defect lesion indicating thrombus.

Interestingly, OCT images of the calcified thrombus presented a low-signal-intensity mass with an overlying signal-rich band. Such OCT findings are usually seen in lesions with a red thrombus,<sup>3)</sup> thin-cap fibroatheroma,<sup>4)</sup> or calcified nodule.<sup>5)</sup> The precise reasons why calcified thrombi present such images are unclear. One possible explanation is the deposition of multiple microcalcifications in a thrombus. Recently, Fujii, *et al* report-



**Figure 2.** Panels **A**, **B**, and **C** show intracoronary images of an angiographic filling defect lesion. **A:** Intravascular ultrasound (IVUS) showed a hypoechoic mass (asterisks) with superficial hyperechoic signal. **B:** Optical coherence tomography (OCT) identified a mass protruding into the lumen with high OCT-signal backscattering and attenuation (asterisks). **C:** Angioscopy revealed a white mass with irregular surface. Arrows indicate stent struts. Panels **D**, **E**, and **F** show IVUS, OCT, and angioscopic images of an angiographically normal site in stents. The majority of the stent struts were uncovered by neointima. Panels **G**, **H**, and **I** show IVUS, OCT, and angioscopic images after aspiration at the site corresponding to **A**, **B**, and **C**, respectively. A mass protruding into the lumen decreased dramatically.



**Figure 3.** Histopathology of aspirated mass. Large amounts of microcalcification were detected in the thrombus by hematoxylin and eosin staining (panel **A**, purple-colored area) and von Kossa staining (panel **B**, black-colored area) (scale bar = 50 μm).

ed that fibrous plaque with large amounts of microcalcifications appeared as low-signal-intensity regions with diffuse borders in OCT.<sup>6)</sup> They suggested that OCT light signal attenuation caused by multiple scattering may be a reason for this appearance. Because OCT measures the intensity of light returning from within a tissue, tissue having a higher heterogeneity of optical index of refraction, such as microcalcification deposition on the luminal surface, may exhibit stronger optical scattering.<sup>6)</sup> On the other hand, it is unclear why angioscopic images of calcified thrombus showed a white material with irregular surface protrusion into the lumen, which resembled white thrombus. One possible reason for this may be the existence of fibrinous materials in a calcified thrombus because a previous study reported that fibrin clots show white color on angiography.<sup>7)</sup>

Regarding the mechanism of the calcified thrombus formation in this patient, we believe that the uncovered struts of multiple overlapping drug-eluting stents might have caused thrombus formation, and that maintenance hemodialysis due to end-stage renal disease could induce calcium deposition in such a thrombus, during which fibrin might have had more affinity for calcium than platelets or red blood cells. But the exact mechanism is still unknown.

Another remaining question in this case is the cause of the frequent stent thrombosis. Premature antiplatelet therapy discontinuation, renal failure, bifurcation lesions, diabetes, and low ejection fraction have been reported as predictors of stent thrombosis after implantation of drug-eluting stents.<sup>8)</sup> Among those factors, the present case was complicated by renal failure and diabetes. In addition, high platelet reactivity to clopidogrel or aspirin has been reported to be associated with an increased risk of stent thrombosis and is a predictor of recurrent ischemic events in patients with acute coronary syndrome.<sup>9)</sup> Recently, on-treatment platelet reactivity can be evaluated by a point-of-care assay using VerifyNow. Aspirin resistance was defined as ARU > 550,<sup>10)</sup> and clopidogrel resistance was defined as PRU > 230.<sup>11)</sup> In the present case, the patients achieved on-aspirin reactivity of 465 ARU and on-clopidogrel reactivity of 153 PRU, which means that platelets were inhibited well by aspirin and clopidogrel. Moreover, not only platelet aggregation activity but also in-stent luminal characteristics might cause late ST.<sup>12)</sup> Drug-eluting stents consist of metal struts, an anti-proliferative drug, and solvent polymer. The drug is slowly released from the polymer, mainly into the perivascular tissue where it suppresses cell proliferation. Indeed, IVUS, OCT, and angiography in the present case revealed that the majority of stent struts were uncovered by neointima, which might be thrombogenic. The solvent polymer remains on the stent struts after drug release and can cause chronic inflammation in the vessel wall, which also leads to delayed arterial healing.<sup>13,14)</sup> In the present case, multiple overlapping drug-eluting stents might have induced these effects more strongly.

In conclusion, we have presented a case with an in-stent

calcified thrombus with diverse *in vivo* images among IVUS, OCT, and angiography. Further histopathological understanding of calcified thrombi visualized by these imaging modalities will be needed, which may provide insight into the mechanism.

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