The role of SCUBE1 in the pathogenesis of no-reflow phenomenon presenting with ST segment elevation myocardial infarction

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Abstract

Objective: SCUBE1 [signal peptide-CUB (complement C1r/C1 s)-EGF (epidermal growth factor)-like domain-containing protein 1] might function as a novel platelet-endothelial adhesion molecule and play pathological roles in cardiovascular biology. Acute myocardial infarction is one of the most common causes of death in modern society. The concept of "no reflow" (NR) refers to a state of myocardial tissue hypoperfusion in the presence of a patent epicardial coronary artery. The main mechanisms of this phenomenon are thought to be high platelet activity and much thrombus burden. So, we researched the role of SCUBE1 in the pathogenesis of NR.

Methods: A total of 142 patients with ST elevation myocardial infarction (STEMI) (n=42 with NR and n=100 without NR) and 50 healthy individuals were prospectively case-control recruited between March 2015 and October 2016 from our outpatient clinics of cardiology department. Patients with STEMI were diagnosed according to American Heart Association (AHA) guideline for the management of STEMI.

Results: The mean SCUBE1 levels of the control subjects were 34±8.4 ng/mL, the mean SCUBE1 levels of patients with STEMI who were treated successfully with primary percutaneous coronary intervention (PCI) were 51±6.2, and the mean SCUBE1 levels of patients with STEMI who had NR phenomenon after primary PCI procedure were 97.2±8.9 ng/mL.

Conclusion: In our opinion, SCUBE1 might contribute to NR phenomenon via thrombus activation and aggregation. The pathophysiology of NR phenomenon is unclear. The present study is the first clinical study that demonstrated that serum SCUBE1 level was significantly higher in patients with NR and that serum SCUBE1 was an independent predictor for the presence of NR in our study population.

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Keywords: no-reflow, platelet activation, SCUBE1

Introduction

There is an important link between platelets and inflammation, thrombosis, vascular, and tissue repair mechanisms. Proliferative, mitogenic, and inflammatory substances are released by activated platelets into the local microenvironment of vascular lesions (1). So, vascular and tissue repair mechanisms are controlled and affected by these activated platelets.

The epidermal growth factor (EGF) superfamily is a group of growth factors, cytokine-like mediators, and extracellular matrix proteins. A new gene for an EGF-related protein was isolated 6 y ago in developing mice (2). This new mammalian gene encodes a protein with a signal peptide at the amino-terminus followed by several EGF-like repeats and one CUB-domain at the carboxyl terminus. This gene family was termed SCUBE, for signal-peptide-CUB-EGF-like domain-containing proteins.

SCUBE1 expression was detected in platelet-rich thrombi and in human atherosclerotic lesions. Tu et al. (3) described adhesion and aggregation experiments that revealed that recombinant fragments of SCUBE enhanced platelet aggregation and adhesion. Activated and adherent platelets release SCUBE1 and it sticks to subendothelial matrix (4). Thus, SCUBE1 plays pathological roles in atherothrombosis by functioning as a novel platelet-endothelial adhesion molecule (5–7). Platelet activation is the most important step in arterial thrombosis and accordingly responsible for the ischemic complications of acute coronary syndrome (ACS) (8–10).

The most common cause of death is acute myocardial infarction (AMI) in modern society (11). The preferred reperfusion method of treatment in AMI is percutaneous coronary intervention (PCI) compared to thrombolytic therapy because of the superior patency rates in the target coronary artery (12). Although the results for PCI in AMI are enough to achieve the desired patency of the target coronary artery, the major drawback is that

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the optimal perfusion is not achieved at the myocardial tissue level in some patients (13). This so called "no-reflow (NR)" phenomenon leaves myocardial tissue hypoperfusion despite the restoration of epicardial coronary artery patency following PCI (14). The main mechanism of this phenomenon is thought to be high platelet activation (15). High platelet activity is a risk factor for thrombotic events. For this reason, in this study, we researched the role of SCUBE1 in the pathogenesis of NR.

Methods

Study design/recruitment and exclusion criteria

In this prospective case-control study, a total of 142 patients with STEMI (n=42 with NR and n=100 without NR) and 50 healthy individuals were prospectively recruited between March 2015 and October 2016 from our outpatient clinics of cardiology department. Patients with STEMI were diagnosed according to American Heart Association (AHA) guideline for the management of ST elevation myocardial infarction (16). NR is defined as a state of myocardial tissue hypoperfusion in the presence of a patent epicardial coronary artery. Baseline clinical demographic characteristics of study groups were reviewed.

Exclusion criteria included significant valvular heart disease, idiopathic cardiomyopathies, any malignancy, rheumatologic and hematologic disease, renal failure (serum creatinine level >2.5 mg/dL), or liver disease (elevated aminotransferases).

This study was approved by the institutional review boards of our Sivas Numune Hospital, and written informed consents were obtained from the participants. The investigation conformed with the principles outlined in the Declaration of Helsinki.

This study was approved by Institutional Ethics Committee of Cumhuriyet University.

Plasma SCUBE1 assays

A full blood count was performed, and routine biochemistry and cardiac enzymes were investigated from blood specimens that were collected from the patient and control groups. In addition, 2 cc of blood was collected in citrate tubes from the control group and the study group at admission to our hospital (before primary PCI procedure and anti-platelet therapy) for investigation. Specimens were centrifuged for 15 min in a 4000-cycle centrifuge device at +4°C. Serum (1 cL3) was placed in an Eppendorf tube, and these were kept at –80°C until the assay was performed. Twenty-four hours before the SCUBE1 evaluation, the Eppendorf tubes were removed and placed in a +4°C environment. Sera were thawed gradually over 24 h, and SCUBE1 levels were measured after the samples had reached room temperature. An enzyme-linked immunosorbent assay kit was used to determine SCUBE1 levels according to the manufacturer's instructions. Specimen absorbance was determined on a VERSA max tunable microplate reader (Molecular Devices, Sunnyvale, CA, USA) at a wavelength of 450 nm. Results were expressed as ng/mL.

Stent implantation procedure

Drug eluting stents (zotarolimus-eluting) were deployed to treat total lesions with standard techniques. During the procedure, patients received heparin to maintain an activated clotting time of ≥250 s. Heparin was not continued after the coronary stenting procedure. Before the stenting procedure, 180 mg of ticagrelor was orally administered to all the study patients. If the thrombus burden was very high, we supplied intravenous glycoprotein IIb/IIIa inhibitor therapy.

Statistical analyses

SPSS (SPSS Inc., Chicago, IL, USA) was used for statistical analyses. The data were evaluated using descriptive techniques (mean, standard deviation). In addition, for quantitative data, the independent samples t test (independent sampling t test) was used to compare groups of parameters that exhibited normal distribution, and the paired t test was used to determine the significance of the differences between the two matched groups. Categorical data were expressed as number and percentage. Pearson's chi-square test was used in the analysis of categorical data. Mann–Whitney U test was used for comparison of nonparametric variables. We calculated that enrolment of 142 patients with STEMI (n=42 with NR, n=100 without NR) in the study group and 50 individuals in the control group would provide a 99.5 power to demonstrate a significant difference in SCUBE1 levels between patients with NR, without NR, and control subjects. Receiver operating characteristic curve (ROC) analysis was used for determining the optimum cut-off SCUBE1 value for predicting NR after primary PCI in patients with STEMI. The independent predictors for the presence of NR were analyzed by using logistic regression analysis. Possible confounding factors were tested in univariable regression analysis and confounders with a p value <0.25 were tested in multivariable logistic regression analysis. A probability value of less than 0.05 was considered the minimum level of statistical significance.

Results

Baseline characteristics and laboratory parameters of patients and healthy control subjects are shown in Table 1. There were no significant differences between groups in terms of age, gender, hypertension, diabetes, smoking, serum creatinine levels, and platelet count. There was no acetylsalicylic acid (ASA) or clopidogrel use in the control and study subjects. Total cholesterol and low-density lipoprotein (LDL) levels did not differ between the patient groups with STEMI but control subjects had lower total cholesterol and LDL levels (p=0.01). The Gensini score of the STEMI with NR group was 34±8.8 and that of the STEMI with successful primary PCI group was 36±7.2. The severity of coronary atherosclerosis was not different between the STEMI groups (p=0.42). The average stent length in the STEMI with NR group was 30±6.2 mm, and the average stent length in the STEMI with successful primary PCI group was

Table 1. Baseline characteristics and laboratory measurements of the study population

Parameters	Control subjects (n=50), mean±SD or median (min–max)	STEMI with successful primary PCI (n=100), mean±SD or median (min–max)	STEMI with NR (n=42), mean±SD or median (min–max)	*P*
Age, y	62±6.4	60±3.3	64±4.7	NS
Sex, female/male	24/26	48/52	19/23	NS
Body mass index, kg/m^2	28.0±4.2	29.6±3.9	30.1±4.4	NS
Glucose, mg/dL	88.5±5.4	90.3±6.5	92.4±3.7	NS
BUN, mg/dL	12.0 (9–19)	14.0 (6–26)	14.0 (9–29)	NS
Creatinine, mg/dL	0.7 (0.5–1.1)	0.7 (0.5–1.2)	0.8 (0.5–1.4)	NS
Uric acid, mg/dL	7.7±1.0	7.5±1.1	7.4±1.3	NS
Total cholesterol, mg/dL	230 (124–312)	288 (196–374)	280 (190–380)	<0.05
Triglyceride, mg/dL	163.7±100.2	181.4±60.3	181.4±61.5	<0.05
HDL, mg/dL	45.3±11.1	46.5±11.6	44.5±11.2	NS
LDL, mg/dL	141.2±43.5	170.4±27.3	168.4±21.9	<0.05
hs-CRP, mg/dL	0.3 (0–0.8)	0.4 (0–2.0)	0.4 (0–2.0)	NS
Hemoglobin, g/dL	13.4±1.0	13.1±1.0	13.7±2.0	NS
Platelet, $\times 10^3$/μL	247.4±54.0	253.9±52.5	250.9±47.5	NS
DM (n)	14	29	15	NS
HT (n)	15	30	16	NS
Smoking (n)	26	49	27	NS
Gensini score, mean	–	34±8.8	36±7.2	NS
Stent length, mm, mean	–	29±4.8	30±6.2	NS
Bifurcation stenting	–	–	–	

BM - body mass index; BUN - blood urea nitrogen; HDL - high-density lypoprotein; LDL - low dencity lypoprotein; NR - no-reflow; NS - not significant; STEMI - ST elevated myocardial infarction; PCI - percutaneous coronary intervention. Statistical significance was set at $P<0.05$. Compatibility of data with normal distribution was investigated using the Kolmogorov–Smirnov test. The t test was used in the comparison of normally distributed data, and the Mann–Whitney U test was used for nonnormally distributed data. The χ^2 test was used to analyze demographic data, and the Pearson correlation analysis was used for correlation analysis

29±4.8. There were no bifurcation stents in any groups. We used glycoprotein IIb/IIIa inhibitors for the STEMI with NR group after PCI procedure. We obtained thrombolysis in myocardial infarction (TIMI) 1 flow at 28 patients in this group. After that, we performed thrombus aspiration for the remaining patients. We obtained TIMI 1 flow at eight patients after thrombus aspiration. We used intracoronary adenosine for the remaining patients, but we could not provide distal coronary blood flow in the target artery. In the remaining subjects, five of them were dead because of hemodynamic instability.

The mean SCUBE1 level of the control subjects was 34±8.4 ng/mL, the mean SCUBE1 level of patients with STEMI who were treated successfully with primary PCI was 51±6.2, and the mean SCUBE1 level of patients with STEMI who had NR phenomenon after primary PCI procedure was 97.2±8.9 ng/mL. The mean SCUBE1 level of patients with STEMI was significantly higher than that of control subjects (p<0.01). The mean SCUBE1 level of patients with STEMI who had NR phenomenon after primary PCI procedure was significantly higher than that of patients with STEMI who were treated successfully with primary PCI (p<0.01) (Fig. 1). The mean SCUBE1 value of patients with NR who died after PCI procedure was 108.4±8.1 ng/mL. The mean SCUBE1 value of that patients was higher than other patients in study group with NR, but this relationship was not significant (p=0.09).

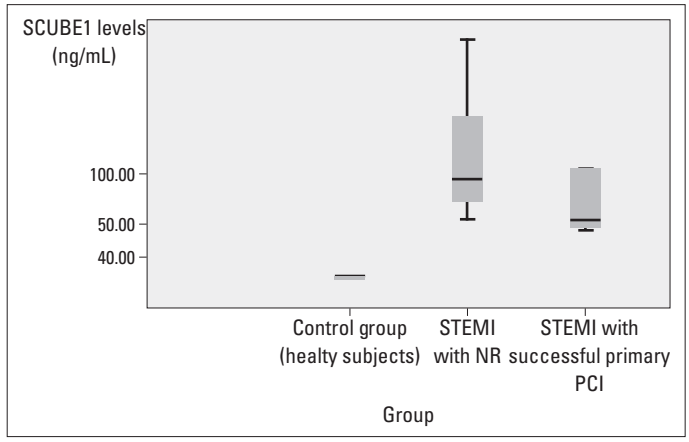

Figure 1. Comparison of SCUBE1 levels between the groups

To determine independent predictors for the presence of NR, univariable and multivariable logistic regression analysis were

Table 2. Univariable and multivariable logistic regression analysis representing the independent predictors of NR phenomenon

	Univariable		Multivariable	
Variables	**OR (95% CI)**	***P***	**OR (95% CI)**	***P***
Age	1.027 (0.991–1.064)	0.140	1.018 (0.977–1.062)	0.388
Hemoglobin	0.831 (0.631–1.094)	0.187	0.940 (0.666–1.328)	0.727
Platelet	1.004 (0.999–1.008)	0.141	1.001 (0.995–1.007)	0.672
LDL cholesterol	1.013 (1.004–1.022)	0.006	1.007 (0.997–1.018)	0.173
hs-CRP	1.183 (0.943–1.484)	0.146	1.039 (0.774–1.394)	0.800
SCUBE1	**1.029 (1.019–1.039)**	**<0.001**	**1.022 (1.011–1.033)**	**<0.001**

CI - confidence interval; hs-CRP - high-sensitivity C-reactive protein; LDL - low-density lipoprotein; OR - odds ratio

performed (Table 2). Univariate regression analysis showed that age, hemoglobin, platelet, LDL cholesterol, high-sensitivity C-reactive protein, and SCUBE1 levels were possible confounding factors for the presence of NR. In multivariable regression analysis, serum SCUBE1 [odds ratio (OR): 1.022; 95% confidence interval (CI): 1.011–1.033, p<0.001] remained as independent predictor for the presence of NR. In addition, ROC analysis was used for determining the optimum cut-off SCUBE1 value for predicting NR after primary PCI in patients with STEMI. The area under the curve (AUC) value was calculated as 0.972 (95% CI: 0.95–0.98). The cut-off SCUBE1 value was observed as 79.8 ng/mL with a sensitivity of 94.4% and specificity of 84.8% (Fig. 2).

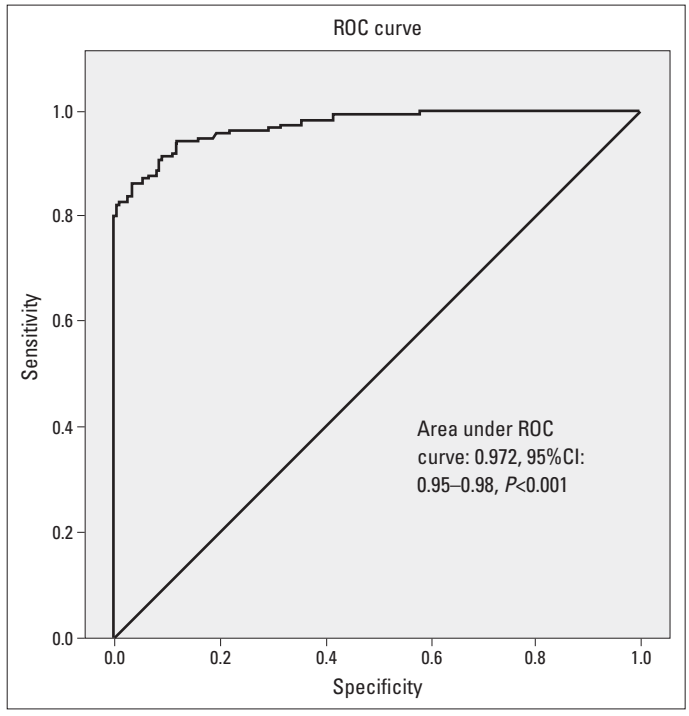

Figure 2. ROC of SCUBE1 and NR phenomenon

Discussion

The major findings of our study were as follows: (i) To the best of our knowledge, the present study is the first clinical study showing a graded relationship between NR development and serum SCUBE1 level. (ii) Furthermore, for the first time, SCUBE1 was found as an independent predictor for the presence of NR.

SCUBE1 is a newly described cell surface molecule that is secreted and expressed throughout early embryogenesis. This protein consists of an N-terminal signal peptide, nine consecutive EGF-like repeats, a spacer region, cysteine-rich repeat motifs, and a CUB domain at the C terminal. These molecules are stored in alpha-granules in inactive platelets. After activation by thrombin, these molecules migrate to the platelet surface. In this stage, small and soluble particles of SCUBE1 play important roles by activating platelets in cardiovascular biology (4). SCUBE1 deposition has been determined immunohistochemically in the subendothelial matrix in advanced atherosclerotic lesions in humans. EGF-like repeats are responsible for adhesive interactions (4). Platelet activation and aggregation are well-recognized as primary reactions in arterial thrombosis and, accordingly, are responsible for the ischemic complications of ACS (6, 7). ACS is initiated by plaque rupture or erosion, with subsequent platelet activation and thrombus formation. The importance of platelet activation in the pathogenesis of this atherothrombotic complication was definitely shown by the application of antiplatelet therapy as the most important management in ACS (9, 10).

NR phenomenon occurs in up to half of the patients after primary PCI and is associated with poor long-term outcome (17). The most important role in the pathogenesis of NR is played by microvascular plugging because of a plaque fragment embolism (18) as suggested by recent studies that have shown the association between the occurrence of NR and plaque characteristics or to platelet-platelet and platelet-leukocyte aggregates (19). Of note, their degree of activation depends on the duration of coronary occlusion. Indeed, Xu et al. (19) found that the transfusion of platelets from mice subjected to a prolonged coronary ischemia exacerbate myocardial reperfusion injury in mice subjected to a brief coronary occlusion. Along with cellular determinants of reperfusion injury, the release of vasoactive substances by platelets plays an important role in inducing sustained vasoconstriction, which further sluggish microvascular coronary flow (20). Thromboxane A2 (TXA2) is the most important vasoactive substance that is released by platelets. Indeed, TXA2 is a po-

tent platelet agonist and coronary vasoconstrictor. Furthermore, it stimulates platelet-leukocyte aggregation by inducing the expression of adhesion molecules on cellular surface (21, 22). In particular, a recent study by Huczek et al. (23) has found that patients with higher platelet activity assessed through PFA-100 (platelet function assay) have a higher percentage, statistically significant, of NR assessed both by angiographic and electrocardiographic parameters. Indeed, the results of this study suggest that platelet activity might also provide early prognostic information about left ventricular performance and adverse clinical events after STEMI.

High platelet activity is a risk factor for thrombotic events. Dai et al. (24) demonstrated the role of SCUBE1 in patients with ACS. According to Dai et al. (24), the soluble form of SCUBE1 might play pathological roles via platelet activation and aggregation. They found that plasma SCUBE1 levels were significantly higher in patients with ACS and acute large-vessel atherothrombotic (LAT) stroke. LAT stroke but not small lacunar stroke had elevated plasma SCUBE1 concentration in the subgroup analysis of acute ischemic stroke. While small lacunar stroke involved the occlusion of small penetrating artery due to lipohyalinosis from sustained hypertension, with less evidence of platelet activation, LAT stroke involved acute platelet activation (25). According to their post-hoc analysis, subjects with acute LAT stroke had significantly higher plasma SCUBE1 than those with transient ischemic attack (TIA), indicating a potential application in the clinical setting to distinguish between acute LAT stroke and TIA. Moreover, plasma SCUBE1 was independently correlated with plasma sCD40L, a marker of platelet activation as well as inflammation. These findings support that plasma SCUBE1 is a biomarker of platelet activation during acute thrombotic complications, such as in ACS and acute LAT stroke.

Our study results are consistent with the findings from previous study. We demonstrated for the first time a graded relation between serum SCUBE1 level and NR development. In multivariable regression analysis, serum SCUBE1 level was found as a significant and independent predictor for the presence of NR.

Study limitations

Our study population was small; there is a need for studies with larger sample sizes.

Conclusion

In our opinion, SCUBE1 might contribute to NR phenomenon via thrombus activation and aggregation. Based on the findings of our study, there is difference in the serum SCUBE1 concentration among studied groups. The present study is the first clinical study that demonstrated that serum SCUBE1 level was significantly higher in patients with NR and that serum SCUBE1 was an independent predictor for the presence of NR in our study population. Also, SCUBE1 levels may predict the development of NR after primary PCI in patients with ACS. More prospective studies are needed to reveal the effect of SCUBE1 levels on the development of NR.

Conflict of interest: None declared.

Peer-review: Externally peer-reviewed.

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References

1. Gawaz M, Langer H, May AE. Platelets in inflammation and atherogenesis. J Clin Invest 2005; 115: 3378-84. [CrossRef]
2. Wu BT, Su YH, Tsai MT, Wasserman SM, Topper JN, Yang RB. A novel secreted, cell surface glycoprotein containing multiple epidermal growth factor-like repeats and one CUB domain is highly expressed in primary osteoblasts and bones. J Biol Chem 2004; 279: 37485-90. [CrossRef]
3. Tu CF, Su YH, Huang YN, Tsai MT, Li LT, Chen YL, et al. Localization and characterization of a novel secreted protein SCUBE1 in human platelets. Cardiovasc Res 2006;71:486–95. [CrossRef]
4. Balzar M, Briaire-de Bruijn IH, Rees-Bakker HA, Prins FA, Helfrich W, de Leij L, et al. Epidermal growth factor-like repeats mediate lateral and reciprocal interactions of Ep-CAM molecules in homophilic adhesions. Mol Cell Biol 2001; 21: 2570-80. [CrossRef]
5. Lindemann S, Gawaz M. SCUBE1-A new scoop in vascular biology? Cardiovasc Res 2006; 71: 414-5. [CrossRef]
6. Türkmen S, Eryiğit U, Karaca Y, Menteşe A, Sümer UA, Yuluğ E, et al. Diagnostic value of plasma signal peptide-Cub-Egf domain-containing protein-1(SCUBE-1) in an experimental model of acute ischemic stroke. Am J Emerg Med 2015; 33: 262-5. [CrossRef]
7. Türkmen S, Şahin A, Günaydın M. The value of signal peptide-CUB-EGF domain-containing protein-1 (SCUBE-1) in the diagnosis of pulmonary embolism: A preliminary study. Acad Emerg Med 2015; 22: 922-6. [CrossRef]
8. Heeschen C, Dimmeler S, Hamm CW, van den Brand MJ, Boersma E, Zeiher AM, et al. Soluble CD40 ligand in acute coronary syndromes. N Engl J Med 2003; 348: 1104-11. [CrossRef]
9. Özkan G, Ulusoy S, Menteşe A, Karahan SC, Cansız M. New marker of platelet activation, SCUBE1, Is elevated in hypertensive patients. Am J Hypertens 2013; 26: 748-53. [CrossRef]
10. Karabacak M, Yiğit M, Türkdoğan KA, Yiğit E, Selek S. Is signal peptide-CUB-EGF domain-containing protein1 a diagnostic biomarker in patients with hypertensive crises. Clin Hemorheol microcirc 2015; 61: 513-22. [CrossRef]
11. Antman EM, Anbe DT, Armstrong PW, Bates ER, Green LA, Hand M, et al. ACC/AHA guidelines for the management of patients with ST-elevation myocardial infarction: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Committee to Revise the 1999 Guidelines for the Management of Patients with Acute Myocardial Infarction) Circulation 2004; 110: 82-292. [CrossRef]
12. Smith SC Jr, Feldman TE, Hirshfeld JW Jr, Jacobs AK, Kern MJ, King SB 3rd, et al. ACC/AHA/SCAI 2005 guideline update for percu-

taneous coronary intervention: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (ACC/AHA/SCAI Writing Committee to Update 2001 Guidelines for Percutaneous Coronary Intervention) Circulation 2006; 113: 166-286. [CrossRef]
13. Michaels AD, Gibson CM, Barron HV. Microvascular dysfunction in acute myocardial infarction: focus on the roles of platelet and inflammatory mediators in the no-reflow phenomenon. Am J Cardiol 2000; 85: 50-60. [CrossRef]
14. Eeckhout E, Kern MJ. The coronary no-reflow phenomenon: a review of mechanisms and therapies. Eur Heart J 2001; 22: 729-39.
15. Jaffe R, Charron T, Puley G, Dick A, Strauss BH. Microvascular obstruction and the no-reflow phenomenon after PCI. Circulation 2008; 117: 3152-6. [CrossRef]
16. Patrick T, O'Gara. 2013 ACCF/AHA guideline fort he management of ST elevation myocardial infarction. Circulation 2013;127:362-425.
17. Rezkalla SH, Kloner RA. No-reflow phenomenon. Circulation 2002; 105: 656-62. [CrossRef]
18. Kusama I, Hibi K, Kosuge M, Nozawa N, Ozaki H, Yano H, et al. Impact of plaque rupture on infarct size in ST-segment elevation anterior acute myocardial infarction. J Am Coll Cardiol 2007; 50: 1230-7. [CrossRef]
19. Xu Y, Huo Y, Toufektsian MC, Ramos SI, Ma Y, Tejani AD, et al. Activated platelets contribute importantly to myocardial reperfusion injury. Am J Physiol Heart Circ Physiol 2006; 290: 692-9. [CrossRef]
20. Frossard M, Fuchs I, Leitner JM, Hsieh K, Vlcek M, Losert H, et al. Platelet function predicts myocardial damage in patients with acute myocardial infarction. Circulation 2004; 110: 1392-7. [CrossRef]
21. Patrono C, Garcı´a Rodrı´guez LA, Landolfi R, Baigent C. Low-dose aspirin for the prevention of atherothrombosis. N Engl J Med 2005; 353: 2373-83. [CrossRef]
22. FitzGerald GA. Mechanisms of platelet activation: thromboxane A2 as an amplifying signal for other agonists. Am J Cardiol 1991; 68: 11-5.
23. Huczek Z, Filipiak KJ, Kochman J, Piatkowski R, Grabowski M, Roik M, et al. Baseline platelet reactivity in acute myocardial infarction treated with primary angioplasty- influence on myocardial reperfusion, left ventricular performance, and clinical events. Am Heart J 2007; 154: 62-70. [CrossRef]
24. Dai DF1, Thajeb P, Tu CF, Chiang FT, Chen CH, Yang RB, et al. Plasma concentration of SCUBE1, a novel platelet protein, is elevated in patients with acute coronary syndrome and ischemic stroke. J Am Coll Cardiol 2008; 51: 2173-80. [CrossRef]
25. Gustafsson F. Hypertensive arteriolar necrosis revisited. Blood Press 1997; 6: 71-7. [CrossRef]

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