Supplementary material

Sadowski M, Ząbczyk M, Undas A. Low-grade lipopolysaccharide-related endotoxemia alters coronary thrombus composition and fibrin clot properties in patients with acute ST-segment elevation myocardial infarction. Pol Heart J. 2024.

Table S1. Patient clinical characteristics, coronary thrombus composition and laboratory findings

Variable	Pre-PCI group, n = 16			Post-PCI group, n = 24		
	LPS <36.1 pg/ml, n = 9	**LPS >36.1 pg/ml, n = 7**	***P*-value**	**LPS <35.9 pg/ml, n = 11**	**LPS >35.9 pg/ml, n = 13**	***P*-value**
Risk factors and clinical presentation						
Age, years	66.5 (12.1)	68.0 (12.3)	0.91	64.7 (12.4)	66.2 (12.1)	0.97
Female sex, n (%)	3 (33.3)	4 (57.1)	0.62	3 (27.3)	5 (38.5)	0.57
Smokers, n (%)	3 (33.3)	3 (42.9)	0.84	4 (36.3)	7 (53.8)	0.4
Type 2 diabetes, n (%)	2 (22.2)	1 (14.3)	0.53	3 (27.3)	2 (15.4)	0.48
Hypertension, n (%)	5 (55.5)	4 (57.1)	0.62	6 (54.5)	8 (61.5)	0.73
Dyslipidemia, n (%)	4 (44.4)	2 (28.6)	0.32	3 (27.3)	6 (46.1)	0.35
BMI, kg/m^2	25.9 (2.5)	27.6 (2.2)	0.34	28.2 (3.4)	26.4 (4.7)	0.45
Time to PCI, h	3.5 (3–8)	6.5 (4.75–33)	0.24	6 (4.5–8.5)	13 (5–72)	0.12
LVEF, %	40.0 (38.7–44.3)	47.5 (38.7–51.2)	0.87	50 (42.5–51)	55 (45–60)	0.42
Thrombus composition						
Fibrin, %	33.7 (27.4–40.4)	42.8 (35.3–57.2)	0.17	25.8 (19.7–46.6)	45.5 (27.9–63.0)	0.04
Platelets, %	10.1 (1.7–37.5)	3.1 (0.9–13.2)	0.4	6.5 (2.3–2.9)	6.5 (2.9–17.9)	0.4
Red blood cells, %	14.4 (7.9–23.8)	10.2 (4.5–22.0)	0.53	30.7 (27.4–41.6)	4.9 (1.3–30)	0.03
White blood cells, %	0.8 (0.5–1.5)	1.4 (0.2–2.9)	0.75	1.16 (0.9–4.7)	1.5 (0.6–5.7)	0.83
Laboratory data						
Hemoglobin, g/l	12.8 (1.5)	12.4 (1.6)	0.71	14.1 (1.9)	13.8 (2.0)	0.98
RBC, 10^{12}/l	4.5 (0.5)	4.4 (0.5)	0.83	4.7 (0.8)	4.9 (0.7)	0.88
WBC, 10^9/l	8.0 (7.4–9.7)	11.3 (8.8–14.3)	0,12	9.9 (8.3–10.9)	9.6 (8.9–10.4)	0.66
Platelets, 10^9/l	263 (229–291)	236 (182–302)	0.52	186 (172–226)	190 (178–241)	0.38
hs-cTnT, ng/l	108 (38.5–211)	623 (244–1315)	0.03	186 (85–1221)	305 (127–1693)	0.97
Creatinine, mg/dl	1.03 (0.15)	1.07 (0.28)	0.83	1.08 (0.16)	1.04 (0.11)	0.9
Fibrinogen, g/l	4.2 (1.3)	4.58 (1.8)	0.63	4.1 (1.1)	4.4 (1.3)	0.73
hs-CRP, mg/l	8.3 (4.0–28.7)	33.0 (24.1–61.6)	0.2	9.3 (4.4–27.3)	13.3 (4.5–41.5)	0.37

MPO, µg/ml	1.0 (0.4)	0.9 (0.6)	0.46	1.0 (0.6)	1.3 (0.6)	0.25
PAI-1 Ag, ng/ml	66.2 (18.6)	57.3 (28.1)	0.35	59.5 (20)	61.9 (23.1)	0.14
TAFI, %	107.7 (11.4)	96.5 (12.4)	0.12	106.7 (14.4)	103.3 (12)	0.7
t-PA, ng/ml	9.6 (1.3)	10.5 (2.1)	0.31	10.7 (1.1)	9.9 (1.6)	0.2
vWF Ag, IU/ml	126.4 (40.1)	145.6 (63.8)	0.75	141.2 (41.6)	155.9 (58.1)	0.52
sCD40L, ng/ml	1.1 (0.2)	1.2 (0.7)	0.22	1.43 (0.52)	1.33 (0.59)	0.35
P-selectin, ng/ml	189.8 (29.3)	198.8 (31.6)	0.46	185 (27.8)	205 (28.9)	0.09
K_s-Thr, $10^{-9}cm^2$	4.3 (1.0)	3.8 (0.9)	0.6	5.7 (1.9)	6.9 (2.3)	0.39
K_s-TF, $10^{-9}cm^2$	4.6 (3.2–5.1)	2.9 (2.7–3.0)	0.3	4.1 (1.0)	5.4 (2.4)	0.2
CLT, min	60 (49–61)	85 (84–96)	0.02	73 (65–82)	77 (67–83)	0.74

Data presented as numbers (percentages), mean (standard deviation), or median (interquartile range), as appropriate

Abbreviations: Ag, antigen; BMI, body mass index; CAD, coronary artery disease; CLT, clot lysis time; CRP, C-reactive protein; cTnT, cardiac troponin T; hs, high sensitive; K_s, permeation coefficient; LVEF, left ventricular ejection fraction; MPO, myeloperoxidase; PAI-1, plasminogen activator inhibitor type-1; PCI, percutaneous coronary intervention; sCD40L, soluble CD40 ligand; TAFI, thrombin activatable fibrinolysis inhibitor; TF, tissue factor; Thr, thrombin; t-PA, tissue plasminogen activator; vWF, von Willebrand factor

Scanning electron microscopy

High-definition images (3500× magnification) were obtained using scanning electron microscopy (JEOL, Tokyo, Japan) in 10 different areas depending on the thrombus size. Each image was divided into 400 squares (Photo Filter 7 software). Finally, 128 ICT images in the pre-PCI and 195 images in the post-PCI group were obtained for further analyses.

Laboratory tests

Plasma fibrin clot permeability (K_s) and clot lysis time (CLT), together with serum LPS, platelet and endothelial activation, fibrinolysis, and inflammation markers, were measured *ex vivo* in 16 patients on admission (pre-PCI group) and in 24 patients on the next morning (post-PCI group). Enzyme-linked immunosorbent assays (ELISAs) were used to determine serum MPO (Calbiochem Millipore, Billerica, MA, US), PAI-1 and tissue plasminogen activator (t-PA; both, Hyphen BioMed, Neuville-Sur-Oise, France), thrombin activatable fibrinolysis inhibitor (TAFI; Chromogenix, Lexington, MA, US), and 2 platelet markers, i.e., soluble CD40 ligand (sCD40L), and P-selectin (both, R&D Systems, Minneapolis, MN, US). vWF antigen (vWF Ag) was measured by latex immunoassay on a STAR coagulation instrument (Diagnostica Stago, Asnieres, France). Serum LPS concentrations were assessed using the ELISA (Cusabio, Houston, TX, US). Clot permeation was assessed using a pressure driven system, with calculation of a permeation coefficient (K_s) using the tissue factor (TF)- and thrombin-based

(Thr) assays. Fibrinolysis induced by recombinant tPA (Boehringer Ingelheim, Ingelheim, Germany), expressed as clot lysis time (CLT), was measured using a TF-induced clot lysis assay [7].