

Bivalirudin infusion to reduce ventricular infarction: the open-label, randomised Bivalirudin Infusion for Ventricular Infarction Limitation (BIVAL) study



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KEYWORDS

- adjunctive pharmacology
- clinical trials
- magnetic resonance imaging (MRI)
- other imaging modalities
- ST-elevation myocardial infarction (STEMI)

Abstract

Aims: The aim of the study was to investigate whether bivalirudin versus unfractionated heparin (UFH) reduces infarct size (IS) for primary percutaneous coronary intervention (PPCI) in large acute myocardial infarction (AMI).

Methods and results: This multicentre open-label trial randomised 78 patients undergoing PPCI for large AMI to bivalirudin or UFH. The primary endpoint was IS, assessed by cardiac magnetic resonance (CMR) five days after PPCI. Secondary endpoints included index of microcirculatory resistance (IMR), CMR-assessed microvascular obstruction (MVO) and ejection fraction, and biomarkers for thrombin activity and cell injury. No difference was observed in mean IS at five days (25.0±19.7 g for bivalirudin vs. 27.1±20.7 g for UFH; p=0.75). Early MVO was numerically lower with bivalirudin (5.3±5.8 g vs. 7.7±6.3 g; p=0.17), with no significant difference in ejection fraction at 90 days (54.6±12.0% vs. 49.1±12.1%; p=0.11). In the biomarkers, thrombin-antithrombin complexes were reduced by 4.8 ug/L over the first day for bivalirudin versus an increase of 1.9 ug/L in the heparin arm (p=0.0003). Acute IMR was lower (43.5±21.6 vs. 68.7±35.8 mmHg×s, respectively; p=0.014). In a planned interim analysis, an approximate 11% reduction in IS was observed with bivalirudin; the trial was discontinued for futility.

Conclusions: This study did not achieve its primary endpoint of significant infarct size reduction in PPCI by prolonged bivalirudin infusion compared to UFH, even though complete thrombin inhibition was achieved in the acute phase, with a lower myocardial microcirculation resistance at the end of the procedure.

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54 Abbreviations

55	AMI	acute myocardial infarction
56	APPROACH	Alberta Provincial Project for Outcome Assessment in Coronary Heart Disease
57		
58	BARC	Bleeding Academic Research Consortium
59	BIVAL	Bivalirudin Infusion for Ventricular InfArction Limitation
60		
61	CMR	cardiac magnetic resonance
62	DTPA	diethylenetriamine penta-acetic acid
63	EDV	end-diastolic volume
64	IMR	index of microcirculatory resistance
65	IQR	interquartile range
66	IS	infarct size
67	ITT	intention-to-treat
68	LVEF	left ventricular ejection fraction
69	MPO	myeloperoxidase
70	MSI	myocardial salvage index
71	MVO	microvascular obstruction
72	PPCI	primary percutaneous coronary intervention
73	SD	standard deviation
74	STEMI	ST-segment elevation myocardial infarction
75	TAT	thrombin-antithrombin complexes
76	TIMI	Thrombolysis In Myocardial Infarction
77	UFH	unfractionated heparin

78 Introduction

79 Primary percutaneous coronary intervention (PPCI) for
80 ST-elevation myocardial infarction (STEMI) reduces infarct size
81 (IS) and improves survival. Even with rapid restoration of epicar-
82 dial flow, final IS is considerable, with a significant contribution
83 from reperfusion injury and microvascular obstruction (MVO) due
84 to oedema and capillary obstruction by microthrombi. Final IS
85 depends on several factors including the occlusion location, dura-
86 tion of occlusion, the burst of thrombin triggered by plaque rup-
87 ture and coronary intervention involving thrombus manipulation¹.

88 Beyond its role in coagulation, thrombin is proinflamma-
89 tory and is implicated in reperfusion injury. On a cellular level,
90 thrombin activates platelets, endothelial cells and leucocytes; con-
91 sequently, it exerts a potent proinflammatory action, with a throm-
92 bogenic and deleterious effect on the microcirculation especially
93 downstream from the culprit lesion². The formation of monocyte-
94 platelet aggregates increases within minutes after PCI, reaching
95 a peak within 2-4 hours². Thrombin is formed and released from
96 the occluding thrombus and by the intra-arterial manipulations of
97 vessel reopening during and after completion of PPCI². Bivalirudin
98 directly inhibits both clot-bound and unbound thrombin, whereas
99 heparin is less effective against clot-bound thrombin². Thrombin
100 inhibition by a prolonged 4-hour bivalirudin infusion after PPCI
101 may therefore reduce inflammation and reperfusion injury.

102 The primary objective of the Bivalirudin Infusion for
103 Ventricular InfArction Limitation (BIVAL) study was to investi-
104 gate whether treatment with bivalirudin infusion for four hours
105 versus unfractionated heparin (UFH) for PPCI in large STEMI

reduces reperfusion injury and improves myocardial healing, resulting in a smaller final IS. For this we used cardiac magnetic resonance (CMR) imaging, the gold standard for measuring IS³, which can also provide a reliable quantitative measurement of global left ventricular parameters and identify prolonged MVO. In a subgroup, we measured invasively the index of microcirculatory resistance (IMR) at the end of the procedure, which predicts clinical outcomes⁴. Finally, we assessed the direct and indirect effects of bivalirudin and UFH on coagulation, platelets, and leucocytes to explore the effect of thrombin inhibition on IS.

Methods

BIVAL was a multicentre, open-label, prospective, randomised controlled trial conducted at four sites in the Netherlands and France in patients undergoing PPCI for a large myocardial infarction.

Adults with STEMI who presented >20 minutes and <12 hours after symptom onset were eligible if they fulfilled the following angiographic criteria: Thrombolysis In Myocardial Infarction (TIMI) 0 or 1 flow in the infarct-related artery (IRA); angiographic score ≥ 21 (sizeable infarction, based on initial angiogram) according to the APPROACH score⁵; and eligible for PPCI. Patients with a history of Q-wave myocardial infarction or who had received antithrombotic therapy other than UFH at first medical contact were excluded. Use of glycoprotein IIb/IIIa inhibitors was permitted only as bail-out therapy for the treatment of no-reflow phenomenon or giant thrombus, defined as >2 times the diameter of the vessel. The full study criteria are detailed online (<https://clinicaltrials.gov: NCT02565147>).

All patients provided written informed consent. The sponsor (The Medicines Company, Parsippany, NJ, USA) was responsible for funding, protocol development in collaboration with the executive committee and the clinical coordinating centre, on-site monitoring and safety surveillance, statistical analyses, and data management. The institutional review board at each site approved the study protocol and activities.

STUDY MEDICATIONS

All patients received, as soon as logistically possible, aspirin at an initial dose of 150-325 mg orally (or 250-500 mg intravenously) and a loading dose of a P2Y₁₂ inhibitor. Administration of UFH at first medical contact or before the angiogram was allowed as per usual practice.

Randomisation (1:1) was carried out using an interactive voice/web response system. All patients randomised to the bivalirudin arm received a bolus of 0.75 mg/kg and an infusion of 1.75 mg/kg/hr for the duration of the procedure and for four hours after completion of PPCI to maintain consistent thrombin inhibition, as (during STEMI) thrombin exerts activity beyond the restoration of flow in the IRA. Patients randomised to UFH were administered a dose as per standard institutional practice. In cases where activated clotting time was used to inform UFH dosing, a target value of ≥ 250 seconds was recommended. Randomisation was stratified by duration of symptom onset to randomisation (<6 hrs versus ≥ 6 hrs) and site.

PRIMARY AND SECONDARY OUTCOMES

The primary outcome was the difference in IS in the bivalirudin versus UFH arm, assessed by CMR performed five days (defined as 5 days \pm 72 hours from randomisation) after PPCI. Key secondary outcomes were the difference between bivalirudin and UFH in other CMR-derived variables of myocardial recovery five days after PPCI, namely LVEF and MVO. LVEF and IS were also assessed by CMR at 90 days. Other secondary outcomes were markers of thrombin activity and cell injury, including thrombin-antithrombin complexes (TAT), platelet microparticles, and myeloperoxidase (MPO), measured by blood sample collection at baseline and three time points after PPCI.

STUDY MEASUREMENTS

In brief, CMR imaging was performed on 1.5-Tesla clinical scanners⁶. Repeated breath holds and gating by electrocardiogram were applied to minimise the influence of cardiac and respiratory motion on data collection. Cine magnetic resonance imaging was performed using steady-state free-precession techniques by a combination of short-axis and three long-axis views (4-chamber, 2-chamber and 3-chamber).

Early MVO was assessed on first-pass perfusion imaging and was performed at rest during 30-50 consecutive heartbeats immediately after injection of gadolinium diethylenetriamine penta-acetic acid (DTPA) (0.1 mmol/kg at 3 mL/s into an antecubital vein followed by 15 mL of saline at 3 mL/s)^{7,8}. Delayed enhancement imaging was performed with a gated breath hold T1-sensitive recovery gradient-echo sequence with a minimum of 10 minutes after infusion of gadolinium DTPA (total of 0.2 mmol/kg i.v.)³. The inversion time was selected to null out the myocardial muscle signal. Typical inversion times ranged from 200 to 300 ms.

Left ventricular function and late gadolinium enhancement were analysed in an independent core lab (CERC) on a workstation using dedicated software. CAAS MRV, 3.4 (Pie Medical Imaging, 2011, Maastricht, the Netherlands) was used for the evaluation of left ventricular function IS and MVO.

Left ventricular end-diastolic and end-systolic volumes were short axis-based and calculated using a combination of long- and short-axis images and are expressed in millilitres⁶. Epicardial and endocardial contours were automatically detected, and manual corrections applied if needed, with consensus between the two readers.

IMR was measured in three of the four preselected sites using a previously described thermodilution technique^{9,10}. IMR was defined as distal coronary pressure multiplied by the mean hyperaemic transit time (expressed in mmHg \times seconds [s]). The results are presented as mean \pm standard deviation (SD) for both treatment arms. In addition, a cut-off point of 32 (associated with better outcomes⁹) was selected as the threshold to categorise IMR.

Biomarker blood sample collection was carried out before study drug administration, at the end of PPCI, at the end of 4-hour infusion, and at 12-24 hours after PPCI on citrate and on ethylenediaminetetraacetic acid (EDTA). The following variables

were measured: TAT and change in TAT (which expresses the net effect of thrombin generation and its inhibition), myeloperoxidase, and microparticles. Care was maximum to respect the preanalytical requirements strictly. Within two hours of blood collection, platelet-free plasma aliquots were prepared by double centrifugation. Plasma aliquots were frozen to -80°C and shipped to the core laboratory (<3 months) for measurement of biomarkers. TAT and myeloperoxidase were measured by an ELISA method, commercialised by Siemens Healthcare Diagnostics, Duisburg, Germany, and ALPCO Diagnostics, Salem, NH, USA, respectively. Platelet microparticles were measured on a flow cytometer (Becton Dickinson, Franklin Lakes, NJ, USA) using annexin V (Beckman Coulter, Brea, CA, USA) and two antibodies (anti-CD41a and anti-CD42b from Becton) and calibration microbeads (Megamix[®]; Biocytex, Marseille, France). Anti-Xa activity was measured in all patients at baseline, and at subsequent time points in heparin-treated patients only, by an amidolytic method with chromogenic substrate (Stachrom[®] Heparin; Diagnostica Stago, Asnières sur Seine, France); direct thrombin inhibition was measured in bivalirudin-treated patients using a modified thrombin time Hemoclot[®] (HYPHEN Biomed, Neuville-sur-Oise, France). Both coagulation tests were performed on a coagulation analyser (STA-R; Diagnostica Stago). All biomarker analyses were carried out by a centralised, independent core laboratory.

Net adverse clinical events, defined as death, reinfarction, ischaemia-driven revascularisation, and Bleeding Academic Research Consortium (BARC) ≥ 3 bleeding were assessed up to five days or at discharge from hospital, whichever came first. Patients were followed to day 90.

Statistical analysis

Continuous variables are summarised as mean \pm SD or as median (interquartile range). Comparisons were made with a Wilcoxon rank-sum test. Categorical variables are presented as frequencies and percentages and were compared with the chi-square test.

The intention-to-treat (ITT) population was defined as all enrolled subjects. The per-protocol population was defined as enrolled subjects who underwent successful PPCI (i.e., final TIMI flow 2 or 3), had CMR data at five days, and were without major protocol deviations. Primary and secondary endpoint analyses are based on the per-protocol population.

Assuming a 23% reduction in IS with bivalirudin (24 \pm 17 g) versus UFH (31 \pm 17 g), and a two-sided type I error rate of 0.05, 190 subjects (95 per arm) would achieve >80% power. Therefore, approximately 200 subjects were planned to be enrolled in the trial to achieve 190 evaluable subjects. A planned interim analysis was performed after enrolment of approximately one third of randomised patients. If the difference in IS between treatments was $\geq 18\%$ at this point the trial would continue and if <18% the study would stop for futility. Statistical analyses were performed using SAS software, version 9.2 (SAS Institute Inc., Cary, NC, USA).

Results

The interim analysis showed an approximate 11% reduction in IS with bivalirudin, and the trial was discontinued for expected futility. At that point, 78 patients had been enrolled (ITT population), 69 of whom completed the study; 64 patients met the inclusion criteria for the per-protocol population (**Figure 1**).

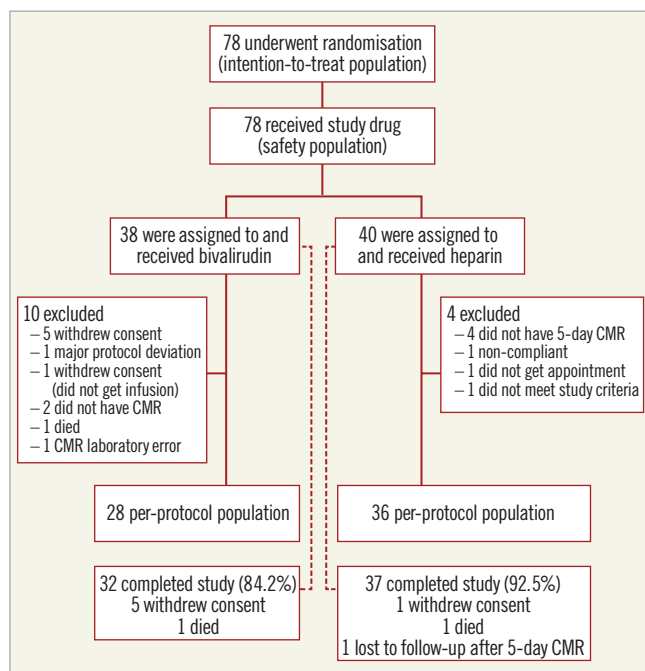


Figure 1. Consort diagram.

Most of the patients (83%) were screened within six hours of symptom onset, and 56% within three hours. Sixty-two of 64 patients received UFH before randomisation (at first medical contact or before the diagnostic angiogram). Mean age was 62.8±11.8 years and 81% were men (**Table 1**). Aspirin was given to 98% of patients and a P2Y₁₂ inhibitor loading dose in 97%.

Radial access was used in 94% of patients (**Table 2**). At presentation, 81% of patients had TIMI 0 flow. The culprit artery was the left anterior descending in 45% of the patients. Thrombus aspiration was performed in 33%; 9% of patients received bail-out glycoprotein IIb/IIIa inhibitor treatment. Mean APPROACH score was 29.8±8.7. Total infusion duration for bivalirudin was 4.09 hours and the total dose was 648 mg, given as a bolus and followed by an infusion. Post randomisation UFH was administered at 58.5 U/kg. No between-group differences were observed for the angiographic findings (**Table 3**).

CMR RESULTS

Overall mean IS at five days was 26.2±20.1 g (16.3±11.6% of left ventricular mass), early MVO was 6.6±6.1 g, and ejection fraction was 48.6±10.9%. CMR data per treatment arm are presented in **Table 4**. No statistically significant reduction was observed in the mean IS at five days (primary endpoint) (25.0±19.7 for bivalirudin

Table 1. Demographic characteristics (per-protocol population) and antiplatelets started before PCI.

Variable	Total (N=64)	Bivalirudin (N=28)	UFH (N=36)
Age (years)	62.8±11.8	62.9±10.6	62.8±12.8
Men, n (%)	52 (81)	22 (79)	30 (83)
Weight (kg)	79.9±13.9	81.0±13.4	79.1±14.5
Previous myocardial infarction, n (%)	4 (6)	0	4 (11)
Previous cerebrovascular event, n (%)	1 (2)	0	1 (3)
Diabetes mellitus, n (%)	4 (6)	0	4 (11)
Smoking status, n (%)			
Current (<30 days)	26 (41)	13 (46)	13 (36)
Former	16 (25)	8 (29)	8 (22)
Never	18 (28)	4 (14)	14 (39)*
Unknown	4 (6)	3 (11)	1 (2.8)
Hypertension, n (%)	24 (38)	9 (32)	15 (42)
Hyperlipidaemia, n (%)	20 (31)	7 (25)	13 (36)
Antiplatelets started before PCI			
Aspirin	63 (98)	27 (96)	36 (100)
P2Y ₁₂ inhibitor	62 (97)	26 (93)	36 (100)
Ticagrelor	47 (76)	17 (65)	30 (83)
Prasugrel	13 (21)	9 (35)	4 (11) [†]
Clopidogrel	2 (3)	0	2 (6)

Data are presented as mean±standard deviation or n (%). *p=0.03; [†]p=0.02. All others were not significant. PCI: percutaneous coronary intervention; UFH: unfractionated heparin

vs. 27.1±20.7 for UFH; p=0.75) (**Figure 2**). Early MVO was numerically lower in the bivalirudin arm (5.3±5.8 g vs. 7.7±6.3 g, p=0.17). There was no significant difference in ejection fraction (p=0.97).

Table 2. Procedural characteristics (per-protocol population)*.

Variable	Total (N=64)	Bivalirudin (N=28)	UFH (N=36)
Radial approach, n (%)	60 (94)	27 (96)	33 (92)
APPROACH score	29.8±8.7	29.1±8.0	30.4±9.3
Pre-PPCI, n (%)			
TIMI flow 0	52 (81)	25 (89)	27 (75)
TIMI flow 1	12 (19)	3 (11)	9 (25)
Thrombus aspiration, n (%)	21 (33)	8 (29)	13 (36)
Balloon predilation, n (%)	20 (31)	7 (25)	13 (36)
Stent, n (%)	64 (100)	28 (100)	36 (100)
Infarct-related artery, n (%)			
Right coronary artery	25 (39)	14 (50)	11 (31)
Left anterior descending	29 (45)	11 (39)	18 (50)
Left circumflex	11 (17)	4 (14)	7 (19)
Bail-out glycoprotein IIb/IIIa inhibitor, n (%)	6 (9)	3 (11)	3 (8)

Data presented as mean±standard deviation or n (%). *All comparisons were p>0.05. PPCI: primary percutaneous coronary intervention; TIMI: Thrombolysis In Myocardial Infarction; UFH: unfractionated heparin

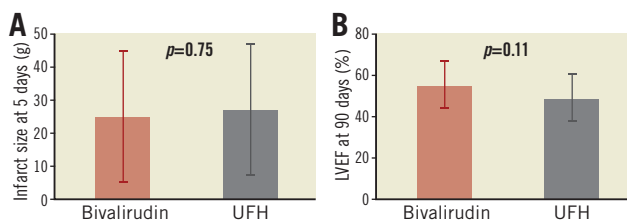
Table 3. Angiographic results at the end of PCI and IMR results.

Variable	Bivalirudin (N=28)	UFH (N=36)	p-value
Angiographic results (per protocol population)			
TIMI flow	2.8±0.4	2.8±0.4	0.91*
Myocardial blush grade	1.8±1.2	1.5±1.2	0.34*
TIMI frame count	27.5±17.4	28.5±13.3	0.40*
IMR results (IMR ITT population)			
Mean hyperaemic distal coronary pressure (mmHg)	66.9±10.6	70.2±18.2	0.33*
Mean transit time (s)	0.7±0.3	1.0±0.5	0.017*
Mean IMR (mmHgxs)	43.5±21.6	68.7±35.8	0.014*
IMR, mmHgxs, n/N (%)			0.11*
<32	9/25 (36)	4/25 (16)	
≥32	16/25 (64)	21/25 (84)	
Data are presented as mean±standard deviation or n (%). *Wilcoxon rank-sum test. †chi-square test. IMR: index of microcirculatory resistance; ITT: intention to treat; UFH: unfractionated heparin			

Data for 90-day CMR were available in 51 patients (Table 4). Overall mean ejection fraction increased to 54.6±12.0 from 49.7±10.5 (change from day 5 to day 90 with bivalirudin: 4.9±11.8%; p=0.066; change with UFH: 1.0±6.9%, p=0.43). No statistically significant difference was observed in ejection fraction at late follow-up (Figure 2). Overall, mean IS decreased independently of the treatment arm (change from day five to day 90 with bivalirudin: -6.0±7.9, p=0.0023; change with UFH: -7.2±12.1, p=0.0038).

Table 4. CMR outcomes up to five days or discharge, whichever came first, and at day 90 after PCI (per-protocol population).

Variable	Bivalirudin N=28	UFH N=36	Difference (%)*	p-value †
CMR outcomes at 5 days				
Infarct size assessed by CMR (g)	25.0±19.7	27.1±20.7	-7.75	0.75
Early MVO (g)	5.3±5.8	7.7±6.3	-31.2	0.17
Late MVO (g)	3.7±5.7	4.2±4.5	-11.9	0.40
LVEF (%)	48.5±11.0	48.6±10.9	-0.2	0.97
EDV (mL)	130.0±40.0	134.5±45.2	-3.3	0.84
ESV (mL)	68.4±28.8	71.1±31.6	-3.8	0.89
CMR outcomes at 90 days				
Infarct size assessed by CMR (g)	18.8±15.5	20.4±18.1	-7.8	0.92
Change in infarct size (g)	-6.0±7.9	-7.2±12.1		0.93
LVEF (%)	54.6±12.0	49.1±12.1	11.2	0.11
Change in LVEF (%)	4.9±11.8	1.0±6.9		0.41
EDV (mL)	142.2±47.3	132.2±36.3	7.6	0.76
ESV (mL)	67.4±37.4	69.4±29.7	-2.9	0.66
Data are presented as mean±standard deviation. *Difference (%) is calculated as 100*(value in bivalirudin group minus value in UFH)/value in UFH. †Wilcoxon rank-sum test. CMR: cardiac magnetic resonance; EDV: end-diastolic volume; ESV: end-systolic volume; LVEF: left ventricular ejection fraction; MVO: microvascular obstruction; UFH: unfractionated heparin				

**Figure 2. Mean±standard deviation. A) Infarct size at day 5. B) Left ventricular ejection fraction (LVEF) at day 90.****INDEX OF MICROCIRCULATORY RESISTANCE**

IMR was measured in 52 patients. Mean transit time was significantly lower in the bivalirudin versus UFH arm (p=0.016) (Table 4). With similar distal coronary artery pressures, this resulted in a lower IMR for bivalirudin (43.5±21.6 vs. 68.7±35.8 mmHgxs, respectively; p=0.014). The percentage of patients with impacted flow in the microcirculation (>32 mmHgxs) was numerically lower with bivalirudin (64% vs. 84%) than with UFH.

BIOLOGICAL MARKERS

Following study drug administration, TAT (indicative of thrombin levels) decreased in the bivalirudin arm and increased slowly in the UFH group (Figure 3A, Table 5). Inhibition of MPO was initially greater in the bivalirudin arm (Figure 3B, Table 5). Significant increases from baseline in platelet microparticles CD41 and CD42 were observed in both groups, with no difference between treatment arms (Figure 3C, Figure 3D, Table 5).

Anti-Xa values measured at admission were similar in both arms, indicating that patients in both groups were similarly

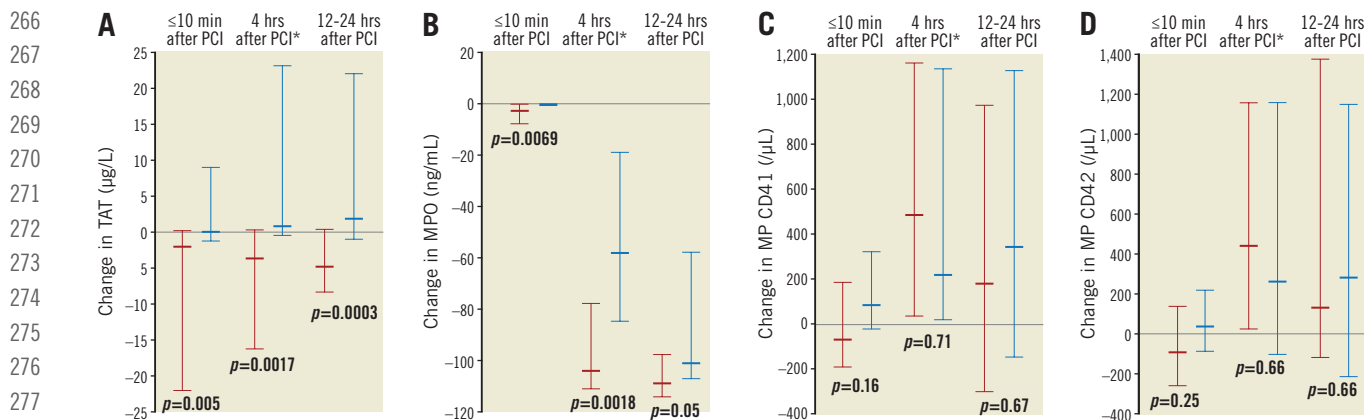


Figure 3. Median (interquartile range) changes from baseline (before study drug administration) to the end of PPCI, the end of 4-hour infusion, and at 12-24 hours after PPCI. A) TAT. B) MPO. C) MP CD41. D) MP CD42. MP: microparticle; MPO: myeloperoxidase; PPCI: percutaneous coronary intervention; TAT: thrombin-antithrombin complexes

pretreated with heparin. After admission, anti-Xa was followed only in the heparin group, showing the expected activity during treatment for the dose administered (Table 5). Similarly, no Hemoclot direct thrombin inhibitor activity was observed

at admission in any patient; direct thrombin inhibitor activity was followed only in bivalirudin-treated patients, showing the expected activity for full thrombin inhibition throughout treatment (Table 5).

Table 5. Biomarker values before study administration and after primary PCI.

Variable	Bivalirudin		UFH		p-value	
	Median	IQR	Median	IQR		
TAT (ug/L)	Before study drug	8.5	(3.90:21.50)	5.1	(3.20:8.10)	0.02
	≤10 min after PCI	5.5	(3.10:8.00)	4.9	(3.40:14.60)	0.61
	4 hrs after PCI	4.3	(3.20:6.50)	5.2	(3.80:30.40)	0.055
	12-24 hrs after PCI	5.2	(3.80:6.80)	8.0	(3.80:25.80)	0.041
MPO (ng/mL)	Before study drug	127.3	(126.40:131.80)	127.3	(125.50:128.10)	0.37
	≤10 min after PCI	126.2	(118.25:130.75)	126.7	(124.20:128.00)	0.65
	4 hrs after PCI	24.2	(16.35:47.20)	63.8	(30.0:107.00)	0.002
	12-24 hrs after PCI	19.7	(15.30:30.55)	26.9	(19.90:58.80)	0.15
MP CD41 (uL)	Before study drug	459.0	(280.0:829.50)	476.0	(184.0:1,002.00)	0.94
	≤10 min after PCI	320.0	(137.0:1,528.00)	479.0	(221.0:1,049.00)	0.43
	4 hrs after PCI	1085.0	(540.0:1,695.00)	807.0	(335.0:1,430.00)	0.60
	12-24 hrs after PCI	645.0	(273.0:2,401.00)	711.5	(372.0:1,754.50)	0.44
MP CD42 (uL)	Before study drug	431.0	(292.0:877.50)	551.0	(208.0:1,033.00)	0.84
	≤10 min after PCI	353.5	(213.0:1,539.00)	507.0	(221.0:1,015.00)	0.52
	4 hrs after PCI	1,125.0	(610.0:1,858.00)	828.5	(344.0:1,690.00)	0.59
	12-24 hrs after PCI	648.0	(309.0:2,518.00)	976.5	(401.50:1,756.50)	0.41
Anti-Xa activity (U/ml)	Before study drug	0.55	(0.36:0.81)	0.66	(0.46:1.13)	0.14
	≤10 min after PCI			1.10	(0.61:1.44)	
	4 hrs after PCI			0.10	(0.05:0.27)	
	12-24 hrs after PCI			0.06	(0.04:0.15)	
Direct thrombin inhibition (ug/ml)	Before study drug	0.12	(0.08:0.17)			
	≤10 min after PCI	7.24	(5.75:11.86)			
	4 hrs after PCI	7.04	(5.99:10.00)			
	12-24 hrs after PCI	0.00	(0.00:0.08)			

IQR: interquartile range; MP: microparticle; MPO: myeloperoxidase; PCI: percutaneous coronary intervention; TAT: thrombin-antithrombin complexes; UFH: unfractionated heparin

CLINICAL OUTCOMES

The rate of in-hospital net adverse clinical events (up to five days or discharge, whichever came first) was 7% (2/28) in the bivalirudin group and 8% (3/36) in the UFH group. BARC ≥ 3 bleeding occurred in 1/28 (4%) patients in the bivalirudin group. One patient (UFH group) died in the period up to 90-day follow-up.

Discussion

Three main observations can be made from this trial. First, the study failed to show a significant reduction in IS at five days with a 4-hour bivalirudin infusion versus standard heparin in PPCI. Second, bivalirudin completely inhibited thrombin generation within 10 minutes of completion of PPCI, whereas UFH did not prevent a small increase in thrombin generation, and this was accompanied by a larger reduction in MPO during bivalirudin infusion. Third, at the end of the procedure, microvascular resistance in the culprit vessel was lower in the bivalirudin-treated patients.

The treatment of myocardial infarction has evolved dramatically over the past 30 years, resulting in a major reduction in mortality. Thrombolysis was the first therapy aimed at reperfusion. Primary angioplasty increased the success rate for restoration of epicardial flow to above 95%. Still, myocardial infarction is a major cause of heart failure related to IS and ventricular remodelling. Additional therapies are needed to take the next step in the reduction of morbidity after acute myocardial infarction. Reperfusion injury and MVO are among the key targets in this process, and bivalirudin has the potential to affect both. In the present study, we failed to demonstrate a significant reduction in IS. The observed approximate 8% smaller mean IS of 25.0 g for bivalirudin versus 27.1 g for UFH was not statistically different in the 64 patients, and nowhere near the anticipated 23% reduction, resulting in the pre-specified early termination of the study. Still, the secondary endpoints that investigated the difference in biological activity of these medications and the microvascular function allow us to generate a hypothesis on the mechanism involved to explain the repeated, but not consistent, finding of a mortality reduction in STEMI patients treated with bivalirudin, which was not explained by the lower incidence of bleeding events¹¹⁻¹³.

Bivalirudin inhibited thrombin more effectively than UFH, as measured by the significant drop in TAT within 10 minutes of injection, which persisted throughout the 4-hour infusion, without an increase observed in UFH-treated patients. As postulated, this may have contributed to the better microcirculation, as expressed by the lower IMR values, in the bivalirudin arm. Even after five days, the CMR data point in the same direction of reduced MVO in the early and late imaging series.

The importance of the coronary microcirculation in determining subject outcomes has been demonstrated previously¹⁴⁻¹⁶. IMR correlates with true microvascular resistance and is independent of the flow in the epicardial arteries. Furthermore, IMR provides a reproducible assessment of the microcirculation, which is independent of haemodynamic perturbations¹⁰.

Both IMR- and CMR-based microcirculatory assessments have been linked to adverse late events⁷, including mortality¹⁷. Although MVO is closely related to IS, a large meta-analysis showed the independent prediction of MVO versus IS on mortality⁸. Potential reasons for cardiac death in patients with MVO have been suggested¹⁸. Patients with, versus those without, no-reflow more often had malignant arrhythmias and early heart failure¹⁸.

MECHANISTIC HYPOTHESIS

Thrombin is a potent thrombogenic agent: it transforms fibrinogen into fibrin, it inhibits fibrinolysis through the thrombin activatable fibrinolysis inhibitor, and it is a potent cell activator². When thrombin activates endothelial cells, it reduces their antithrombotic capability, causes the release of prothrombotic factors (e.g., tissue factor), and modifies cell permeability. The change in permeability results in cell oedema, which (along with microthrombosis) is one of the major factors responsible for MVO. Thrombin is initially generated from the lesion at the time of plaque rupture; then, during restoration of flow in the coronary artery and through thrombus manipulation, more thrombin is released from inside the clot and continues to be generated even after completion of PCI. Among the circulating cells, leucocytes are activated by thrombin and release their granular content, such as plasma MPO. The formation of platelet-leucocyte aggregates indicates the effect of thrombin and is implicated in MVO².

The amount of thrombin generated varies between patients and this, in combination with the small size of this study, explains the significantly different TAT levels at baseline between the groups. After initiation of anticoagulation, TAT levels in the bivalirudin arm were immediately reduced and remained low, whereas an increase was observed in the heparin arm, indicative of the direct effect of bivalirudin on thrombin, expressing a complete inhibition of bound and unbound thrombin.

Plasma MPO values, which were similar at baseline in the two groups, decreased four hours after the procedure, with a greater reduction in the bivalirudin arm. After 24 hours, MPO levels were similarly low in both groups. This response may be because polymorphonuclear leucocytes, in addition to being activated by thrombin, have a direct role in the thrombotic process, known as NETosis¹⁹.

Thrombin inhibition (expressed through TAT measurement) and leucocyte activation (expressed through MPO measurement) were the two parameters (both thrombin-related) that demonstrated fluctuations during the acute phase of the myocardial infarction and which responded to thrombin inhibition, underlining the role of thrombin and its effect on the microcirculation. Platelets, which often attract focus both clinically but also for research reasons, were not differentially involved, as suggested by the platelet microparticle values, which were similarly dispersed throughout the study and reflect the lack of a difference in the antiplatelet treatment between the groups.

Limitations

The small sample size, due to early discontinuation of the trial, is an obvious limitation. The sample size is even smaller due to

372 the predefined condition that the analysis would be performed in
373 the per-protocol population, even though results are consistent for
374 the larger ITT population. Owing to the small size and the early
375 discontinuation, any results should be addressed with caution and
376 should better be accepted as hypothesis-generating. Furthermore,
377 97% of patients received some dose of UFH at first medical con-
378 tact, so any potential benefit of bivalirudin is in addition to UFH.

379 Conclusions

381 Using a 4-hour bivalirudin infusion in the PPCI setting resulted in
382 complete thrombin inhibition in the acute phase, whereas UFH did
383 not prevent a small increase in thrombin generation. This strategy
384 resulted in a lower IMR but failed to reduce IS, the primary end-
385 point of this study. The trial was discontinued for expected futility
386 after a planned interim analysis. The preliminary findings of the
387 secondary endpoints provide a hypothesis for understanding the
388 repeated, but not consistent, finding in previous studies of a mor-
389 tality reduction with bivalirudin.

391 Impact on daily practice

392 The main purpose of anticoagulation at the acute phase of myo-
393 cardiac infarction is inhibition of thrombin. Even though this
394 study failed to demonstrate the effect of prolonged bivalirudin as
395 adjuvant therapy during PPCI to reduce IS versus UFH, and is
396 unlikely to impact on guidelines or clinical practice, observations
397 from the secondary endpoints carry the potential to explain the
398 clinical benefit of complete and prolonged inhibition of thrombin.

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411 Conflict of interest statement

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