Impaired myocardial t-PA release in patients with coronary artery disease

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Aims: Myocardial ischemia remains a significant perioperative complication in coronary artery disease (CAD) patients. We hypothesized that noxious stimuli during major surgery are associated with an acute release of tissue-type plasminogen activator (t-PA) into the coronary circulation, and that this response is reduced by CAD.

Methods and results: Two patient groups, with ($n = 14$) and without ($n = 8$) CAD, were studied during the initial phase of heart surgery. After retrograde great cardiac vein catheterizations during closed-chest conditions, coronary arterial–venous concentration gradients of t-PA and plasminogen activator inhibitor type-1 (PAI-1) were measured together with coronary blood flow measurements, allowing derivation of coronary net release rates. Pre-surgery atrial pacing, performed to evaluate the influence of increases in heart rate (+40 beats/min) and coronary blood

flow (+80 ml/min), did not significantly alter coronary net release of t-PA or PAI-1 in either patient group. Sternotomy induced a prominent increase in coronary net release of both total and active t-PA in the non-CAD group. This response was considerably reduced in the CAD group.

Conclusions: This study provides the first analysis of coronary t-PA release during major surgery and demonstrates a deficient local endothelial t-PA release in patients with CAD. This suggests a reduced local fibrinolytic capacity in CAD patients, which may explain the increased risk for coronary thrombosis in this patient group.

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IN the majority of cases, myocardial infarction and cardiac death are due to coronary artery thrombosis secondary to acute rupture of a coronary atheromatous plaque.¹ Activation of the fibrinolytic system by recombinant tissue-type plasminogen activator (t-PA) is effective in inducing reperfusion in the thrombus-occluded artery by degradation of intravascular fibrin clots. Normal blood vessels have an inherent capacity to acutely release t-PA upon stimulation from an intracellular storage pool in the endothelium.² The observation that products from activated platelets and the coagulation cascade are powerful stimuli for regulated t-PA release suggests that this acute secretory response is a local thrombo-protective mechanism to promote spontaneous dissolution of intravascular thrombi.^{3,4} Existence of such a counter-regulatory feedback might explain the substantial incidence of spontaneous reperfusion in the thrombosed artery in myocardial ischemia (after acute myocardial infarction).⁵

Conversely, if this thrombo-protective response is impaired, the risk of formation of a flow-arresting thrombus may increase. In support of this hypothesis, we have showed that subjects with a genetic low capacity for t-PA release, i.e. t-PA-7, 351T allele carriers,^{6,7} have an increased risk for myocardial infarction.⁸ This finding was recently confirmed in the Framingham Heart Study.⁹

Besides genetic factors, functional impairment of endothelial cells may be expected to alter the capacity for acute t-PA release. We have demonstrated that patients with essential hypertension and chronic renal failure, two well-known risk factors for myocardial infarction, have an impaired capacity for pharmacologically induced t-PA release^{10,11} Similar findings have been reported for obese individuals.¹²

Of particular interest is the coronary circulation, in which history of smoking has been associated with impaired t-PA net release.¹³ The coronary t-PA release responsiveness has also been shown to

correlate inversely to the degree of atheromatous plaque burden.¹⁴

Based on earlier animal experimental findings,^{15,16} we postulated that stress during major surgery would elicit a local fibrinolytic response through an increased regulated release of t-PA. We hypothesized that coronary atherosclerosis might alter this response. The specific aim was therefore to measure coronary net fluxes of t-PA in patients with or without coronary artery disease (CAD) at the stressful events during cardiac surgery. We focused on sternotomy procedures. To explore the role of alterations in coronary blood flow and heart rate (HR) for coronary release patterns of t-PA, atrial pacing was performed before surgery as an additional part of the study protocol.

Methods

Study patients

Twenty-two patients scheduled for heart surgery that included sternotomy were included in the study. We included consecutive patients in two groups: subjects with CAD who underwent coronary bypass surgery and subjects without CAD who underwent other types of open-chest heart surgery.

The study was undertaken after approval from the Umeå University Research Ethics Committee, in accordance with the Declaration of Helsinki (version 2000). All subjects provided informed consent before entry in the study.

Inclusion criteria for CAD patients ($n = 14$) consisted of a history of stable angina pectoris and a coronary angiography showing two to three vessel CAD ($\geq 50\%$ stenosis) including the left anterior descending artery.

Patients in the non-CAD group ($n = 8$) had no history of angina pectoris and underwent a pre-surgery coronary angiography demonstrating no signs of CAD. Indications for surgery were aortic valve insufficiency ($n = 6$) or proximal aortic dilation ($n = 2$). Exclusion criteria were unstable angina, diabetes mellitus, non-sinus rhythm, left or right bundle branch block, pacemaker, manifest heart failure, renal failure requiring dialysis and on-going therapy with anti-coagulants other than acetylsalicylic acid and low molecular heparin. Further, patients with coagulation abnormalities as indicated by history or routine pre-surgery coagulation profile were excluded.

General pre-surgery procedures

Study subjects refrained from salicylate intake and other anti-inflammatory drugs for a week, as well as from β -receptor blockers and ACE inhibitors for 24 h pre-operatively. Pre-medication comprised flunitrazepam 1 mg orally and morphine 0.1 mg/kg intramuscularly. Using local anesthesia, a 20 G radial artery catheter (BD, Becton Dickinson, Stockholm, Sweden) was inserted.

General anesthesia was induced with midazolam 0.06–0.1 mg/kg and fentanyl 0.002–0.005 mg/kg, and maintained with isoflurane. Before endotracheal intubation, pancuronium 0.1 mg/kg was given. Ventilation was adjusted to normocapnia as indicated by end-tidal CO₂ levels (MM206C Gas Monitor, Arthema, Stockholm, Sweden) and intermittent arterial blood gas analyses (ABL 5, Radiometer, Copenhagen, Denmark). A continuous infusion of isotonic acetated solution at a rate of 5 ml/kg/h was administered.

Through the right internal jugular vein a 7/F catheter was inserted in the pulmonary artery and a 7/F 2-thermistor coronary sinus (CS) thermodilution and pacing catheter (CCS-7U-90A, Ball end, Webster Labs, Los Angeles, CA) was placed in the great cardiac vein (GCV). Fluoroscopic guidance was used for positioning of the catheters and the position of the GCV catheter was confirmed using a bolus injection of a contrast solution (Omnipaque®, Nycomed, Oslo, Norway).

Hemodynamic measurements

Measurements included pressures in radial and pulmonary arteries and in the superior vena cava (Siemens Sirecust 1280, Dräger Medical, Lubeck, Germany). Blood flow in the great cardiac vein, Q_{GCV} , was derived using the retrograde thermodilution technique¹⁷ and intracatheter thermal transport correction.¹⁸ Impedance signals were processed by a two-channel Wheatstone bridge (CBA-210, Webster Labs). Measurements were recorded on an eight-channel Graphtec Linearcorder WR 3310 (Western Graphtec Corp., Irvine, CA).

Electrocardiogram (ECG) was used for monitoring of HR, arrhythmias and ST changes (60 ms after the J-point). Five ECG leads were employed during sternotomy (Sirecust 1280, Dräger Medical) and 12 leads during pacing (Mingograph 7, Siemens-Elma AB, Stockholm, Sweden). Computerized vectorcardiography (MIDA 1000, Ortivus Medical AB, Täby, Sweden) with eight electrodes according to the Frank system¹⁹ was used as an additional

means for myocardial ischemia detection. Signals were sampled at 500 Hz, averaged for periods of 15 s in three orthogonal components and projected along three perpendicular axes (X, Y and Z dimensions). ST-segment deviations (spatial ST change vector magnitude, STC-VM) were measured 20 ms after the J-point.¹⁹

Atrial pacing

After a rest period of 20 min under general anesthesia, atrial pacing was performed at closed chest conditions. As further discussed below, this was done to analyze possible effects of HR increases and changes in coronary blood flow on coronary t-PA release. Baseline pacing was done at a frequency of 5–10 beats above each subject's spontaneous HR. The pacing frequency was then increased in steps of 10 beats/min (bpm). Before each increase in pacing rate, a 5–8 min stabilization period was allowed, at the end of which measurements were performed. The maximal pacing rate was pre-set at 140 bpm. As a safety measure, pacing was stopped at the appearance of one of the following signs of myocardial ischemia:

1. If myocardial net lactate production occurred (YSI Sport 2300 Stat Plus; Yellow Springs Instruments Inc., Yellow Springs, OH). This was the single cause for discontinuation of pacing in one subject in the CAD group.
2. If ST changes in at least two leads appeared (>0.1 mV, in a 12-lead ECG). This was the single cause for discontinuation of pacing in two subjects in the CAD group.
3. If STC-VM of more than $100 \mu\text{V}$ was demonstrated.²⁰ This was the single cause for discontinuation of pacing in nine subjects in the CAD group.

In the non-CAD group, pacing was not intentionally discontinued in any patient below 140 bpm, although the highest HR obtainable in two patients turned out to be 130 bpm.

Surgery

Preparation for surgery started 20 min after completion of pacing procedures, when bolus doses of fentanyl of 0.01–0.015 mg/kg, midazolam 0.04–0.05 mg/kg and pancuronium 0.1 mg/kg were administered and the infusion rate of isotonic acetated solution was increased (10 ml/kg/h).

Midline sternotomy was performed approximately 10 min after the start of surgery and required about 10–15 s.

Measurement sequence

Peripheral venous blood samples were obtained for analysis of plasma levels of t-PA and plasminogen activator inhibitor type-1 (PAI-1) at about 14:00 hours the day before surgery (pre-surgery, Table 1), while blood sampling for lipid status and glucose was done during preparation for anesthesia. Blood pressures, ECG and HRs were continuously recorded, while Q_{GCV} was recorded intermittently in conjunction with blood samplings from the

Table 1

Baseline characteristics of study groups.

	CAD (n = 14)	Non-CAD (n = 8)
Sex (male/female)	9/5	8/0
Age (years)	66 (48–76)	51 (45–58)*
BMI	26 (20.4–29.2)	25.5 (22.0–29.5)
SAP (mmHg)	142 $\pm$ 7	152 $\pm$ 7
DAP (mmHg)	75 $\pm$ 3	74 $\pm$ 7
MAP (mmHg)	96 $\pm$ 4	101 $\pm$ 5*
Functional classification (NYHA)		
I	0/14	7/8
II	0/14	1/8
III	14/14	0/8
IV	0/14	0/8
Smoking history		
Non-smoker	10/14	8/8
Smoker	4/14	0/8
ASA within 7 days	0/14	0/8
Lmwh	0/14	0/8
β -blockers	11/14	3/8
Lipid-lowering agents	11/14	1/8*
Long-acting nitrates	9/14	0/8*
ACE-inhibitors	4/14	5/8
Calcium channel blocker	6/14	2/8
S-Triglycerides (mmol/l)	2.16 $\pm$ 1.06	1.37 $\pm$ 0.57
S-Cholesterol (mmol/l)	4.45 $\pm$ 0.85	5.16 $\pm$ 0.74
S-Fibrinogen (g/l)	2.74 $\pm$ 0.44	2.21 $\pm$ 0.52*
S-Hemoglobin (g/l)	136 $\pm$ 10	145 $\pm$ 8
Pre-study venous concentration		
Active t-PA (ng/ml)	1.15 $\pm$ 0.60	0.92 $\pm$ 0.33
Total t-PA (ng/ml)	10.36 $\pm$ 3.10	9.91 $\pm$ 4.68
Total PAI-1 (ng/ml)	31.74 $\pm$ 15.49	23.66 $\pm$ 10.82

*Significant differences ($P < 0.05$) are indicated (CAD group vs. non-CAD group).

BMI, body mass index; HR, heart rate; SAP, systolic arterial blood pressure; DAP, diastolic arterial blood pressure; MAP, mean arterial blood pressure; NYHA, New York Heart Association functional classification; *Smoker*, denotes smoking within the last year; *Non-smoker*, denotes no smoking within the last year; ASA, acetylsalicylic acid; Lmwh, low molecular weight heparin; ACE inhibitors, angiotensin-converting enzyme inhibitors; CAD, coronary artery disease; t-PA, tissue-type plasminogen activator; PAI-1, plasminogen activator inhibitor type 1.

radial artery and the GCV for determinations of t-PA, PAI-1 and lactate. Measurements were performed at resting HR, at baseline pacing, at each pacing step, before skin incision (control), and at 1, 3, 5 and 10 min after the completion of sternotomy.

Analyses of plasma t-PA and PAI-1

Blood samples for the analysis of plasma t-PA and PAI-1 were collected in tubes containing 1/10 of 0.45 M sodium citrate buffer, pH 4.3 (Stabylite[®], Biopool[®] International, Umeå, Sweden), placed on ice and centrifuged for 20 min at 2000 g and 4 °C. The plasma was frozen and stored at –80 °C. Plasma concentrations of total t-PA antigen and total PAI-1 antigen were determined with ELISAs (TintElize[®] t-PA Biopool[®] International and COALIZA[®] PAI-1 Chromogenix, Haemochrom Diagnostica AB, Mölndal, Sweden, respectively). Both assays detect free and complexed forms of the respective proteins with equal efficiency.^{21,22} Active t-PA was analyzed by a biofunctional immunosorbent assay (Chromolize[™] t-PA, Biopool[®] International) and expressed in nanograms per milliliter using the specific activity of 0.60 IU/ng (data on file, Biopool[®] International). Blood samples from each patient were analyzed in duplicate on a single microtiter plate. The intra-assay coefficient of variation was $2.7 \pm 0.2\%$ for total t-PA, $2.4 \pm 0.2\%$ for active t-PA and $3.9 \pm 0.2\%$ for total PAI-1.

Calculations

Great cardiac venous plasma flow (QP_{GCV}) was calculated using Q_{GCV} and arterial hematocrit (hct), as determined in duplicate using a microhematocrit centrifuge according to the following formula:

$$QP_{GCV} = Q_{GCV}((101 - \text{hct})/100)$$

Coronary net release rates of total t-PA, active t-PA and total PAI-1 were calculated as follows, assuming that plasma concentrations of t-PA and PAI-1 are similar in the radial artery and in the coronary arteries,²³

$$\text{Coronary net release rate} = (C_{GCV} - C_A) \times QP_{GCV}$$

where C_{GCV} is the plasma concentration in the GCV and C_A the plasma concentration in the radial artery.

Statistics

When not otherwise stated, data are presented as a mean $\pm$ standard error of the mean. All analyses were performed using SPSS 11 for

Windows (SPSS Inc., Chicago, IL). P -values < 0.05 were considered significant.

For all repeated measures analysis (ANOVA), degrees of freedom were corrected according to the Huynh–Feldt procedure for possible violation of the assumption of sphericity. When significant main or interactive effects were found, pre-defined contrasts within groups (relative to baseline) or between groups were made.

For evaluation of peak coronary net release of total and active t-PA, paired t -test (within group) and independent samples t -test (between groups) were used.

To analyze the probability that calculated net release or uptake indices were different from 0, one sample t -test was used. χ^2 test was used for comparisons between CAD and non-CAD patients as regards patient characteristics.

For the analyses of the association between total PAI-1 and coronary blood flow, the Spearman rank correlation coefficient was used.

Results

Baseline patient characteristics

There was a preponderance of male subjects in both study groups.

CAD patients all demonstrated angiographically verified three-vessel disease. The CAD group was characterized by higher age, more limitations in physical activity and higher pre-study fibrinogen blood levels (Table 1). There was no difference between groups in the pre-study venous concentration of total t-PA, active t-PA or total PAI-1 the day before surgery.

Atrial pacing. By design, this study allowed an analysis of the role of alterations in HR and Q_{GCV} for coronary net release of t-PA. Among data during pacing sequences, we extracted findings from pacing rate levels that produced changes in HR and coronary flow similar to those produced by sternotomy (Table 2). Pacing-induced HR increases up to 40 bpm were not associated with significant changes in pre-surgical coronary net release rate of either total or active t-PA in either group (Table 2). Nor were net release rates of PAI-1 significantly altered (data not shown). Simultaneously, Q_{GCV} was augmented by about 35 ml/min during HR increases of 20 bpm (both groups) and by about 80 ml/min during HR increases of 40 bpm (non-CAD group).

Table 2

Atrial pacing events.

	Control	HR increase +10 bpm	HR increase +20 bpm	HR increase +40 bpm
Number of patients				
CAD	11	11	11	8
Non-CAD	8	8	8	8
HR (bpm)				
CAD	65 ± 2	76 ± 2*	86 ± 2*	102 ± 2*
Non-CAD	78 ± 2	88 ± 2*	98 ± 2*	118 ± 2*
Q_{GCV} (ml/min)				
CAD	129 ± 23	146 ± 22*	145 ± 21*	167 ± 18*
Non-CAD	125 ± 17	149 ± 19*	157 ± 21*	207 ± 30*
ΔQ_{GCV} (ml/min)				
CAD		17	16	38
Non-CAD		24	32	82
Coronary net release of total t-PA (ng/min)				
CAD	3 ± 27	-25 ± 16	-6.5 ± 20	-56 ± 41
Non-CAD	-17 ± 10	56 ± 25	-4.4 ± 54	-20 ± 51
Coronary net release of active t-PA (ng/min)				
CAD	3.6 ± 2	-0.3 ± 1	0.9 ± 1	1.6 ± 1
Non-CAD	-2.5 ± 1	-3 ± 1	-2.4 ± 1	-0.4 ± 1

Q_{GCV} denotes blood flow changes in the great cardiac vein. Data are presented as mean ± standard error of the mean (SEM).

*Significant differences ($P < 0.05$) vs. control.

HR, heart rate; CAD, coronary artery disease; bpm, beats per minute; t-PA, tissue-type plasminogen activator.

Sternotomy

Hemodynamic data. During resting anesthesia, HR was highest in the non-CAD group (Fig. 1). MAP and HR increased rapidly directly after sternotomy in both groups. Q_{GCV} also increased, but significantly more in the non-CAD group. Ten minutes after sternotomy, the mean increase in HR was 21 bpm in the non-CAD group and 12 bpm in the CAD group. Simultaneously, mean Q_{GCV} had increased by 87 ml/min in the non-CAD group and by 23 ml/min in the CAD group (Fig. 1). No ST changes or serious arrhythmias were seen on the five ECG leads employed during sternotomy.

Coronary net release of t-PA. Pre-surgery net release of t-PA was low and not significantly different between groups (Fig. 2). Sternotomy was associated with prominent increases in net release of both total and active t-PA in the non-CAD group, while there were only minimal, although significant, alterations in these variables in the CAD group (Table 3). As there were inter-individual differences in the time profile of the t-PA response during the 10-min period following sternotomy, we performed an additional analysis based on peak net release rates. Mean peak release rates of both total and active t-PA were significantly higher in the non-CAD compared with the CAD group (Fig. 2). There was no significant correlation between age and coronary net release rates in either group.

Coronary net release of PAI-1. To investigate whether differences in PAI-1 contributed to our findings with regards to active t-PA, net release of PAI-1 antigen was determined. The arterial input of PAI-1 into the coronary vasculature (arterial plasma concentration times Q_{GCV}) was similar for the two groups before surgery, but was higher in the non-CAD group after sternotomy (Fig. 3). However, net release of PAI-1 did not change significantly in response to sternotomy in either group (Table 3).

Discussion

Coronary vascular disease is a significant risk factor for perioperative morbidity during major surgery.²⁴ The present study is unique by investigating the specific impact of CAD on coronary t-PA release rates in such a setting. Also, we analyzed the respective roles of increases in HR and myocardial blood flow for the net release of t-PA into the coronary circulation. We believe such factors are important for the understanding of involved mechanisms behind the increased risk.

One limiting circumstance for the conductivity of this study turned out to be the difficulty to enroll controls, i.e. patients without radiographic signs of CAD. Such patients are scarce in our institutional patient mix, and are only very seldom included in studies of this kind in the literature. This adds to the uniqueness of our study.

Fig.1. Heart rate (A) great cardiac vein blood flow (B) and mean arterial pressure (C) at control and at 1, 3, 5 and 10 min following sternotomy in coronary artery disease (CAD) patients (closed squares) and non-CAD patients (open squares). Data presented as mean $\pm$ standard error of the mean. Significant differences ($P < 0.05$) indicated * (vs. control) and # (between groups).

Fig.2. Peak coronary net release of total (A) and active (B) tissue-type plasminogen activator (t-PA) during the 10 min period following sternotomy in coronary artery disease (CAD) patients (closed bars) and non-CAD patients (open bars). Data are presented as mean $\pm$ standard error of the mean. Significant differences ($P < 0.05$) indicated * (vs. control) and # (between groups).

Our key finding is that sternotomy induces a regulated release of t-PA in the human coronary vascular bed and that peak coronary release rates of both total and active t-PA were on the average four times higher in the non-CAD group compared with the CAD group. This observation suggests that coronary atherosclerosis is associated with an impaired capacity to release t-PA, and thus reduced capability to initiate a local thrombo-protective response. The possible clinical relevance of this finding is highlighted by a recent study by Robinson et al.²⁵ They determined forearm net release of t-PA in response substance P infusion in 98 patients with angiographically proven stable coronary heart disease. Lower t-PA release rates were associated with increased incidence of cardiovascular events after 42 months follow-up.

Table 3

t-PA and PAI-1 following sternotomy.

	Control	1 min	3 min	5 min	10 min
Arterial concentration (ng/ml)					
Total tPA					
CAD	10.6 ± 0.75	11.1 ± 0.75*	11.3 ± 0.78*	12.3 ± 0.89*	12.4 ± 0.87*
Non-CAD	9.59 ± 1.11	10.4 ± 1.44*	11.4 ± 1.18*	11.7 ± 1.21*	13 ± 1.6*
Active tPA					
CAD	0.42 ± 0.08	0.54 ± 0.09*	0.65 ± 0.12*	0.66 ± 0.11*	0.7 ± 0.12*
Non-CAD	0.59 ± 0.12	0.87 ± 0.16*	0.92 ± 0.16*	0.94 ± 0.15*	1.18 ± 0.25
Total PAI-1					
CAD	55.7 ± 6.64	56.2 ± 7.53	52.3 ± 6.12	55.4 ± 6.7	53.3 ± 6.26
Non-CAD	41.6 ± 6.7	43.9 ± 7.34	45.2 ± 7.31	46.3 ± 8.28	44.9 ± 9.11
Great cardiac venous concentration (ng/ml)					
Total tPA					
CAD	10.5 ± 0.7	11.4 ± 0.82*	11.2 ± 0.77*	12.3 ± 0.89*	13 ± 0.97*
Non-CAD	9.75 ± 1.06	10.8 ± 1.42	11.8 ± 1.22*	12.6 ± 1.2*	13.5 ± 1.54*
Active tPA					
CAD	0.42 ± 0.08	0.57 ± 0.1*	0.64 ± 0.11*	0.68 ± 0.12*	0.78 ± 0.14*
Non-CAD	0.63 ± 0.12	0.88 ± 0.17*	0.98 ± 0.18*	0.98 ± 0.17*	1.41 ± 0.24*
Total PAI-1					
CAD	59.7 ± 8	58.4 ± 7.72	55.4 ± 6.97	58.3 ± 7.52	59.1 ± 7.72
non-CAD	45.2 ± 7.04	46.2 ± 7.98	44 ± 7.71	47.5 ± 8.82	44.5 ± 9.6
Coronary net release (ng/min)					
Total tPA					
CAD	-5.6 ± 5.9	11.4 ± 9.3	-10.2 ± 14.2	-1.3 ± 11.7	28.1 ± 10.7*
Non-CAD	12.0 ± 9.9	65.4 ± 56.3	45.0 ± 26.1	97.3 ± 28.3*,†	63.5 ± 45.5
Active tPA					
CAD	0.85 ± 1.00	2.17 ± 1.37	-0.88 ± 1.52	1.47 ± 1.58	3.52 ± 2.56
Non-CAD	2.91 ± 1.67	2.76 ± 2.63	9.66 ± 7.32	12.12 ± 7.56	27.15 ± 8.66*,†
Total PAI-1					
CAD	243 ± 178	116 ± 248	587 ± 590	439 ± 416	642 ± 510
Non-CAD	135 ± 65	296 ± 177	-140 ± 170	114 ± 382	9 ± 91

Data are presented as mean ± standard error of the mean (SEM).

*Significant differences ($P < 0.05$) are indicated (vs. control).

†CAD group vs. non-CAD group.

CAD, coronary artery disease; t-PA, tissue-type plasminogen activator; PAI-1, plasminogen activator inhibitor type 1.

Fig. 3. Arterial input of plasminogen activator inhibitor type-1 (PAI-1) into the coronary vasculature (arterial plasma concentration times Q_{GCV}) following sternotomy in coronary artery disease (CAD) patients (closed bars) and non-CAD patients (open bars). Data presented as mean ± standard error of the mean. Significant differences ($P < 0.05$) indicated * (vs. control).

As to possible underlying mechanisms for our findings, tachycardia brought on by increased intrathoracic pressure or intracoronary infusion of isoprenaline induces coronary t-PA release.^{26,27} However, in the present study pre-operative increases in HR by pacing did not produce a coronary t-PA release, suggesting that HR by itself did not contribute to the t-PA response to sternotomy. This interpretation is supported by the observation that during pacing rates up to 200 bpm, both coronary arterial-venous concentration gradients of t-PA in man²⁸ and coronary net release of t-PA in animals²⁹ remain unaltered.

In the coronary circulation, increases in blood flow and t-PA release occur in parallel in some settings.^{14,15,27,30} In the present study, a wide range of changes in coronary flow rates did not influence coronary t-PA release, indicating that local release rates were not strictly blood flow dependent. This is in line with the observation that pharmacological

augmentation of coronary blood flow by nitroprusside or clevidipine does not induce a regulated local release of t-PA.^{14,29}

As shown both in experimental and clinical studies, sympatho-adrenal activation by several different stimuli can induce a rapid regulated release of t-PA.^{29,31,32} As specifically regards the coronary vascular bed, experimental data show a prompt net release of t-PA following selective stimulation of sympathetic cardiac nerves²⁹ or sternotomy.¹⁶ The role of sternotomy as an initiator of sympathetic activation is illustrated by the fast increase in cardiac norepinephrine spillover observed in patients undergoing coronary artery bypass grafting.³³ Whether β -adrenergic activation, generally implicated in the regulation of t-PA,³⁴ is involved also in such mechanisms in the coronary circulation is still unsettled.

Our findings suggest that regulated release of t-PA following surgical stress is reduced in patients with CAD. Although there were differences among study patients as regards HR or coronary blood flow, such circulatory factors could not explain these findings.

The reduced t-PA release response could reflect relative depletion of t-PA at the local storage pool by coronary atherosclerosis or altered release responsiveness not related to access of endothelial t-PA.

The relative influences of these factors still remain to be clarified. Newby et al.¹⁴ reported that coronary release of active t-PA varies inversely with coronary plaque burden in CAD patients after pharmacological stimulation, which speaks in favor of the first hypothesis. However, this association was observed for active t-PA and not for total t-PA. Because plasma levels of PAI-1 correlated negatively with coronary release of active t-PA, it is possible that their finding at least in part was explained by variations in plasma PAI-1. In contrast, in the present study, CAD patients showed a parallel reduction in coronary release of both total and active t-PA. Furthermore, the arterial inflow of PAI-1 to the coronary vascular bed was higher in the non-CAD group than in the CAD group, which demonstrates that the reduced coronary release of active t-PA in the CAD group is not referable to effects of the inhibitor. This is in line with recent findings that, in the stimulated situation, local t-PA protein release, and not PAI-1, is the major determinant of net release of active t-PA.³⁵ Collectively, our data, therefore, illustrate a reduced t-PA protein secretion in the CAD group. Clearly, further studies are needed to elucidate underlying mechanisms for this finding.

There are some possible limitations of this study. Current medications, with potential intrinsic effects on t-PA release, were more common in CAD patients (Table 1). However, organic nitrates do not influence resting plasma levels of t-PA or PAI-1³⁶ and lipid-lowering treatment does not influence pharmacologically-induced regulated release of t-PA.³⁷ Adrenergic β -receptor activation has been implicated in the release of t-PA,³² but selective β -1-receptor blockers were discontinued 24 h before surgery in the current study. Confounding effects by ACE treatment,³⁰ through impaired degradation of bradykinin, are less likely,³⁸ as there were no differences in resting plasma t-PA levels between patient groups. A history of smoking is associated with a reduced t-PA release in response to pharmacological stimulation in the forearm and in the coronary vascular beds.^{14,39} However, in the CAD group the majority (10/14) of patients was non-smoking, and therefore such factors are not likely to explain our findings. There were no women in the non-CAD group, but resting systemic levels of t-PA do not seem to differ between genders.⁴⁰ Finally, and predictably, patients in the CAD group were older compared with the non-CAD group. We could not explain the impairment of t-PA release in CAD patients by this circumstance and actually, forearm desmopressin-induced t-PA release has been shown to *increase* with age.⁴¹

Conclusion

In summary, this controlled study provides the first analysis of coronary t-PA release during major surgery and demonstrates a deficient local endothelial t-PA release in patients with CAD. This finding has several important implications. The first involves the understanding of coronary t-PA kinetics during surgical stress. The second, and most important, involves possible secondary effects of CAD on thrombolytic capacity. We suggest that reduced regulated release of t-PA, associated with coronary atherosclerosis, might increase the risk of coronary thrombotic events during surgery.

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