Fibrin formation and platelet activation in patients with myocardial infarction and normal coronary arteries


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Coronary spasm is the mechanism most often postulated to explain the rare combination of myocardial infarction and angiographically normal coronary arteries, although the reported evidence for its role is circumstantial rather than conclusive. Whereas the importance of thrombosis in myocardial infarction is uncontested in the presence of significant coronary artery disease, there is little in vivo evidence for thrombosis in angiographically normal coronary arteries.

Among 11 consecutive patients with acute myocardial infarction undergoing thrombolytic therapy with recombinant tissue plasminogen activator (rtPA) 3.2 ± 0.7 h after onset of chest pain, and angiography 10.2 ± 4.5 days later, three young men had normal coronary arteries. Their cases are documented electrocardiographically, enzymatically and angiographically. Mean plasma levels of fibrinopeptide A (FPA) and beta-thromboglobulin (BTG) were clearly elevated before and during rtPA therapy: FPA 52 ± 41 ng ml⁻¹, BTG 257 ± 46 ng ml⁻¹. They did not differ significantly from corresponding mean plasma levels in the eight patients with severe coronary artery disease: FPA 67 ± 66 ng ml⁻¹, BTG 181 ± 75 ng ml⁻¹.

We conclude that fibrin formation and platelet activation are probably equally important in the early hours of myocardial infarction, whether or not significant coronary artery disease is present.

Introduction

Myocardial infarction with angiographically normal coronary arteries is a rare event and an enigma that has received much attention in the literature[1-6]. The prevalence of normal or nearly normal coronary arteries is high, however, in certain subsets of patients with myocardial infarction, such as in young women using oral contraceptives and smoking[7-9], in young men during or following severe physical exercise[10,11] or in very young patients[2,12,13]. Suggested mechanisms for the 'unproven combination'[41] of myocardial infarction and normal coronary arteries include: coronary spasm[14-17], coronary emboli[2], thrombosis with spontaneous lysis[18,19], regression of coronary atheromatoses[20,21], trauma[22], small vessel disease[23] and angiographically unidentifiable coronary lesions[24]. Acute myocardial ischaemia without coronary atheromatoses can also originate from an extreme imbalance between oxygen demand and supply or an abnormal haemoglobin-oxygen dissociation[25].

Fibrinopeptide A (FPA), cleaved from fibrinogen by thrombin[25], is a very sensitive marker of thrombosis in vivo[27,28]. Elevated FPA plasma levels have been reported in patients with acute myocardial infarction[29-32]: unstable angina[33] and venous thromboembolism[34]. Similarly, beta-thromboglobulin (BTG) plasma levels reflecting enhanced platelet activation[35] have been shown to be elevated in patients with acute myocardial infarction[36,37] and unstable angina[37,38].

We measured plasma levels of FPA, BTG and platelet factor 4 (PF4) in 11 consecutive patients with acute myocardial infarction who underwent intravenous thrombolytic therapy with rtPA and angiography. Three patients had normal coronary arteries. Their cases are documented here and the markers of fibrin formation and platelet activation compared with those measured in patients with...
myocardial infarction and significant coronary artery disease.

Methods

PATIENTS AND MANAGEMENT

Inclusion and exclusion criteria for thrombolytic therapy were according to the European Cooperative rtPA study except for the time lapse from onset of pain (< 4 h) and anticoagulation that excluded our patients. 100 mg rtPA (predominantly single chain material) provided by Boehringer-Ingelheim, Switzerland, was infused intravenously over 3 h. All patients underwent biplane left ventriculography and selective coronary arteriography between 5 and 19 (mean 10 ± 4·5) days after thrombolytic therapy.

COAGULATION AND PLATELET TESTS

Blood samples were taken by careful venepuncture before starting the rtPA infusion and 90 min later during rtPA therapy, but before initiating the heparin infusion. They were collected into precooled sample tubes, containing the following anticoagulants (for 9 ml of blood): 1 ml 0·1 M citrate, 1 ml cytosin theophyllin adenosin dipyramidol (CTAD) (Boehringer–Mannheim) supplemented with 200 μg D-phenyl-prolyl-arginine-chloromethylketone (PPACK) as thrombin inhibitor. The blood samples were carefully mixed, immediately cooled on ice and centrifuged at 4°C/2500 g for 30 min within 1 hour of sampling. The plasma was stored at — 70°C. A record was kept of each blood sample in order to identify eventual difficulties in sampling and processing.

FPA was determined in our laboratory using a previously published radioimmunoassay (RIA) with the following modifications: cross-reacting fibrinogen was eliminated by bentonite absorption before using the fibrinogen-free supernatant for the RIA. Free antigen was separated from bound antigen by use of an immobilized second goat-anti-rabbit antibody (Immunobeads, Bio Rad laboratories). Previously measured levels of FPA in 15 normal individuals were 1·9 ± 0·8 ng ml⁻¹. BTG and PF4 were determined by specific ELISA, both obtained from Boehringer–Mannheim, F.R.G. Normal values for BTG are 10–40 ng ml⁻¹. These values are comparable to BTG levels obtained by RIA. PF4 plasma levels were measured in order to exclude in vitro platelet activation by venepuncture artefact (normal range: < 10 ng ml⁻¹, a BTG/PF4 ratio < 3 indicating in vitro platelet activation.)

In addition, blood samples for plasma creatine kinase/MB levels were taken 8, 12, 16, 24 and 48 h after the onset of thrombolytic therapy.

Results

PATIENTS WITH NORMAL CORONARY ARTERIES (GROUP A)

The three cases (numbers 5, 9 and 10) with angiographically normal coronary arteries after myocardial infarction are documented separately in Figs 1–3. All blood samples for FPA and BTG measurements were taken without difficulty.

Case 5 (Fig. 1)

A young sportsman, 27 years of age and a regular smoker, had a 4-month history of transient precordial discomfort at rest, but a normal maximal treadmill test, suggesting the possibility of spastic angina. He was admitted with severe nitroglycerin-resistant chest pain that had begun during an ice-hockey match. The electrocardiogram showed an acute inferolateral myocardial infarction. Intravenous thrombolytic therapy with rtPA was initiated 3 h 15 min after the onset of chest pain. A bradycardiac nodal rhythm followed by self-limited ventricular tachycardia/flutter and a sudden relief of chest pain suggested reperfusion 1 h later. Another short episode of ventricular tachycardia 2 h after the end of thrombolytic therapy and a double peaked curve of creatine kinase/MB plasma levels suggested a re-occlusion and secondary reperfusion. No further complication occurred. An inferior Q-wave infarction evolved subsequently. The coronary arteries were normal, except for systolic narrowing of the left anterior descending artery by myocardial bridging. This abnormality, however, could not be responsible for the acute ischaemic event in the inferior myocardial region, nor probably for the history of transient precordial discomfort at rest, since the diastolic diameter of the left anterior descending artery was also normal. FPA plasma levels were elevated (6·3 ng ml⁻¹ and 39·4 ng ml⁻¹), indicating thrombosis before and 90 min after initiation of thrombolytic therapy. The corresponding high BTG plasma levels (172 and 256 ng ml⁻¹) and normal PF4 levels (10 and 29 ng ml⁻¹) documented markedly enhanced platelet activation in vitro without activation.
Figure 1 27-year-old man with acute inferior myocardial infarction undergoing thrombolytic therapy with rtPA 3.3 h after onset of chest pain. See text for details.
Figure 2 43-year-old man with acute anterior myocardial infarction undergoing intravenous thrombolytic therapy with rtPA 3-5 h after onset of chest pain. See text for details.
Figure 3  38-year-old man with acute infero-posterior myocardial infarction undergoing intravenous thrombolytic therapy with rtPA 3-4 h after onset of chest pain. See text for details.
Table 1  Pertinent data on 11 patients with acute myocardial infarction and angiographically normal (group A) or significantly diseased (group B) coronary arteries after thrombolysis

<table>
<thead>
<tr>
<th>Case</th>
<th>Sex</th>
<th>Age (yr)</th>
<th>Time onset pain to TL (h)</th>
<th>ECG signs of MI</th>
<th>CK/MB-peak (hours from onset TL)</th>
<th>Days since TL</th>
<th>CAD Patency IRA</th>
<th>Res. Sten. IRA</th>
<th>Wall motion in jeopard. region</th>
<th>FPA before TL (ng mg⁻¹)</th>
<th>BTG before TL (ng ml⁻¹)</th>
<th>PF4 before TL (ng ml⁻¹)</th>
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**group A (normal coronary arteries)**

| 5    | m   | 27     | 3-3 | inf-lat | 1680/91 (12 h) | 5 | 0 | + (RCA) | 0% | normal LV | 6-3 | 39-4 | 172 | 256 | 10 | 29 |
| 9    | m   | 43     | 3-5 | ant     | 1652/87 (20 h) | 19 | 0 | + (LAD) | 0% | a inf | 73-4 | 119-2 | 253 | 275 | 111 | 153 |
| 10   | m   | 38     | 3-4 | inf-post | 2230/175 (8 h) | 10 | 0 | + (CX) | 0% | a inf | 73-4 | 119-2 | 253 | 275 | 111 | 153 |
| Mean |     | 36     | 3-4 |         | 2259/145 | 11-3 | 51-7 | 257 | 41-4 | 46 |
| ± SD |     | 8-2    | 0-1 |         | 594/47 | 7 |

**group B (significant coronary artery disease)**

| 1    | m   | 50     | 4   | inf-lat | 3255/500 (8 h) | 5 | 1VD | + (RCA) | 55% | a inf-post | 17-9 | ---* | 159 | ---* | 44 | ---* |
| 2    | m   | 35     | 2   | inf-post | 744/87 (12 h) | 9 | 1VD | + (RCA) | 90% | h inf | 235-8 | 63-6 | 287 | 188 | 183 | 103 |
| 3    | m   | 62     | 3-6 | inf-post | 2193/145 (12 h) | 16 | 2VD | + (RCA) | 90% | h-a inf | 30-7 | 155-2 | 100 | 193 | 12 | 42 |
| 4    | m   | 55     | 2-9 | inf-post | 771/83 (16 h) | 8 | 2VD | + (RCA) | 65% | normal LV | 16-8 | 102-1 | 43 | 131 | 3 | 40 |
| 6    | m   | 49     | 4   | inf-post | 1705/175 (12 h) | 8 | 2VD | + (RCA) | 90% | h-a inf | 30-7 | 155-2 | 100 | 193 | 12 | 42 |
| 7    | f   | 64     | 2-7 | ant-lat | 1371/183 (8 h) | 11 | 1VD | + (LAD) | 99% | d ap | 68-9 | 70-0 | 198 | 263 | 67 | 139 |
| 8    | m   | 40     | 3-7 | ant-lat | 5250/442 (8 h) | 14 | 1VD | + (LAD) | 80% | a-d ant-ap | 51-4 | 41-0 | 167 | 102 | 43 | 10 |
| 11   | m   | 49     | 2-2 | ant     | 639/93 (8 h) | 7 | 1VD | + (LAD) | 99% | normal LV | 1-6** | 13-0 | 233 | 289 | 143 | 190 |
| Mean |     | 50-5   | 3-1 |         | 1991/213 | 9-8 | 83% | 66-8 | 181 | 46-7 | 75 |
| ± SD |     | 9-9    | 0-8 |         | 1585/164 | 3-7 | 16% | 65-7 | 75 |

*data eliminated because of spurious elevation by blood sampling difficulty; **heparin pretreatment; a = akinesia, ant = anterior, ap = apical, BTG = beta-thromboglobulin, CAD = coronary artery disease, CK/MB = creatine kinase/MF-fraction, CX = circumflex artery, d = dyskinesia, ECG = electrocardiogram, FPA = fibrinopeptide A, h = hypokinesia, inf = inferior, IRA = infarct-related artery, LAD = left anterior descending artery, lat = lateral, MI = myocardial infarction, PF4 = platelet factor 4, post = posterior, RCA = right coronary artery, Res. Sten. = residual stenosis, TL = thrombolysis, 1,2,3-VD = 1,2,3-vessel disease.
Case 9 (Fig. 2)

A 43-year-old man, a heavy smoker without history of angina, was admitted with severe nitroglycerin-resistant chest pain and electrocardiographic signs of an acute anterior myocardial infarction. Intravenous thrombolytic therapy with rtPA was initiated 3 h 30 min after onset of pain. Ventricular extrasystoles in salvos 50 min later were interpreted as a reperfusion arrhythmia, since chest pain abruptly disappeared. No complication occurred. Peaking in creatine kinase/MB plasma levels was early. An anterior Q-wave infarction, with loss of R-potentials from V1 to V4, developed. Angiography, 19 days after the acute event, showed a localized but distinct apical akinesia, attributable to a long left anterior descending artery. Coronary arteries, however, were normal. FPA plasma levels were elevated (15-3 and 56-4 ng ml\(^{-1}\)) before and during thrombolytic therapy, as were BTG plasma levels (284 and 304 ng ml\(^{-1}\) respectively), indicating thrombosis and platelet activation. Since PF4 levels (199 and 215 ng ml\(^{-1}\)) were also elevated, however, additional platelet activation in vitro had to be presumed in this case.

Case 10 (Fig. 3)

A 38-year-old man with no history of angina was admitted with nitroglycerin-resistant chest pain that had begun during a soccer game. With typical electrocardiographic signs of acute inferoposterior myocardial infarction, intravenous thrombolytic therapy with rtPA was started 3 h 25 min after the onset of chest pain. The pain diminished gradually during therapy. No reperfusion arrhythmia was noted. Creatine-kinase MB plasma levels peaked very early. An inferior Q-wave infarction evolved within 24 h. No complication occurred. Angiography 10 days later confirmed an inferior akinesia corresponding to the posterior descending branch of a dominant circumflex artery. However, except for minor wall irregularities, the coronary arteries were normal. FPA (73-4 and 119-2 ng ml\(^{-1}\)) and BTG (172 and 256 ng ml\(^{-1}\)) plasma levels were markedly elevated, documenting fibrin formation and platelet activation in vivo with some platelet activation in vitro (PF4 levels 111 and 153 ng ml\(^{-1}\)).

Discussion

PREVALENCE OF NORMAL ANGIOGRAPHY AFTER MYOCARDIAL INFARCTION

The prevalence of angiographically normal coronary arteries after myocardial infarction is 1–4% according to bigger series\(^{13,43}\). No significant coronary stenosis or zero vessel disease is found in 7–19% of postinfarction patients\(^{4,13-23}\). Very young patients\(^{13}\), however, especially women taking oral contraceptives and smoking\(^{7-9,19}\) and young men who sustain myocardial infarction during or immediately following severe physical exercise\(^{10,11}\) have high prevalence (30–45%) of angiographically normal or insignificantly diseased coronary arteries.

CORONARY SPASM

Spasm is the mechanism most often postulated to explain the combination of myocardial infarction and angiographically normal coronary arteries\(^{14-17,43}\), although the reported evidence for its
role is circumstantial rather than causal. Among 679 young survivors of myocardial infarction undergoing angiography, Gohlke et al.\(^1\) found 25 patients with normal coronary arteries, 32 with non-significant ( < 50%) lesions and 112 with unifocal disease; none of these patients had a history of repeated angina at rest of the Prinzmetal type prior to myocardial infarction. Oliva et al. demonstrated coronary spasm angiographically in six patients early after acute myocardial infarction\(^4\), but the spasm consistently occurred at the site of a severe coronary lesion, whereas no spasm was demonstrated in two cases of myocardial infarction and normal coronary arteries\(^3\). Vasospasm has been incriminated in myocardial infarction during or immediately following vigorous exercise\(^10\).

The success of early thrombolytic therapy has established the great importance of thrombosis compared with spasm in myocardial infarction. Rentrop et al. demonstrated the negligible effect of intracoronary nitroglycerin very early after the onset of chest pain\(^44\). Vasocclusion, however, might play a role in reocclusion during thrombolytic therapy\(^45\).

Markers of platelet activation do not specifically indicate vasospasm, since aggregating platelets may exert their effect by mechanical obstruction, enhancement of thrombosis or thromboxane A\(_2\)-mediated vasocstriction. Our measured BTG levels document platelet activation in patients with acute MI whether significant coronary artery disease is present or not. Compared with plasma levels measured between 6 and 32 h after the onset of infarction (64 ± 21 ng ml\(^{-1}\)\(^{132}\)), the levels of BTG measured between 2 and 4 h after onset of chest pain were substantially higher. This might reflect a higher platelet activity in the early hours of myocardial infarction. Patients with normal coronary arteries had higher BTG levels than patients with significant coronary disease (257 ± 45 vs 181 ± 75 ng ml\(^{-1}\)). Whether this corresponds to more spastic activity in agreement with Ogasawara et al.\(^46\) remains speculative in view of the small group size in our study.

CORONARY THROMBOSIS

The causal role of thrombosis in acute myocardial infarction and unstable angina with underlying significant coronary artery disease is hardly contested today. In vivo evidence is based on angiography\(^47,48\), angioscopy\(^49\), elevated plasma levels of fibrinopeptide A\(^29-33\) and cross-linked fibrin degradation products\(^50,51\), and the success of early thrombolytic therapy.

To our knowledge, evidence of a direct connection between angiographically normal coronary arteries and in vivo markers of thrombosis has not been provided so far. Elevated plasma levels of fibrinopeptide A measured as early as 3–2 ± 0.7 h after the onset of chest pain in patients with enzymatically and electrocardiographically evolving myocardial infarction suggest a causal role of thrombosis. Since FPA levels in patients with normal coronary arteries after thrombolytic therapy are not different from levels in patients with significant residual coronary disease, it might be assumed that thrombosis was the mechanism of coronary occlusion in both groups. The success of thrombolytic therapy and the failure of previously given nitroglycerin in all patients support this assumption.

On the other hand, one cannot exclude the possibility that myocardial ischaemia or necrosis per se, by mechanisms so far not elucidated, such as endothelial damage in the ischaemic territory, may be responsible for the activation of both coagulation and platelets\(^38\). A small intracoronary thrombus may not be the sole explanation for the high FPA plasma levels measured. Ischaemia would then be not only the consequence but also a possible cause of further fibrin formation. Elevated FPA levels in unstable angina\(^33,38\), in contrast to stable angina\(^37,38\), may either favour the hypothesis that intracoronary thrombosis also plays a major role in transient ischaemic episodes at rest, commonly interpreted as coronary spasm, or indicate that severe ischaemia per se activates coagulation and platelets.

Plasma levels of FPA measured between 2 and 4 h after the onset of chest pain were significantly higher than our previously published levels\(^30,32\), which were measured between 6 and 32 h after the onset of infarction in non-heparinized patients. This might reflect a time-dependent decrease of thrombotic activity in acute myocardial infarction.

Absence of angiographically detectable coronary lesions does not imply a 'normal' coronary artery. Early endothelial cell damage without visible obstruction might have important sequelae, which have been called 'rapid progression of coronary artery disease'\(^60\). Platelet deposition on such an endothelial lesion, leading to intense coronary spasm of sufficient duration to provoke stasis and
finally, thrombosis, is a sequence of events postulated to explain myocardial infarction with angiographically normal coronary arteries. A reduced activity and enhanced inhibition of endogenous tissue plasminogen activator in patients with myocardial infarction without visible coronary artery disease might, however, lead to thrombosis without preceding spasm.

Finally, FPA is not a specific marker of coronary thrombosis, but could also reflect left ventricular, venous or extravascular fibrin generation. However, none of our three young patients with normal coronary arteries had a left ventricular aneurysm, atrial enlargement or fibrillation, signs of venous stasis or a central catheter. The venepuncture was done with extreme caution and blood samples obtained without difficulty. Extracoronary fibrin formation therefore seems improbable.

Conclusion

Myocardial infarction occurs in spite of angiographically normal coronary arteries. Fibrin formation and platelet activation are of crucial importance whether a significantly diseased or an apparently normal coronary artery occludes, as shown by equally elevated plasma levels of fibrinopeptide A very early after the onset of chest pain, the ineffectiveness of nitroglycerin and the efficacy of thrombolytic therapy.

While coronary spasm alone has not been shown conclusively to be responsible for myocardial infarction, an adjunctive role cannot be excluded. High beta-thromboglobulin plasma levels indicate a marked enhancement of platelet activity in the early hours of myocardial infarction, whether significant coronary disease is present or not.

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References


