CORONARY THROMBOLYSIS WITH TISSUE-TYPE PLASMINOGEN ACTIVATOR IN PATIENTS WITH EVOLVING MYOCARDIAL INFARCTION

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Abstract Tissue-type plasminogen activator is a naturally occurring, clot-selective activator of fibrinolysis. We recently reported that human tissue-type plasminogen activator isolated from a Bowes-melanoma–tissue-culture supernate lysed coronary thrombi in dogs without depleting circulating fibrinogen or α_2-antiplasmin, in contrast to the case with streptokinase and urokinase. In the present study coronary thrombolysis, confirmed angiographically, was induced within 19 to 50 minutes with intravenous or intracoronary tissue-type plasminogen activator in six of seven patients with evolving myocardial infarction. Circulating fibrinogen, plasminogen, and α_2-antiplasmin were not depleted by this agent, in contrast to the case in the two patients subsequently given streptokinase. In the one patient in whom lysis was not inducible with tissue-type plasminogen activator, it was also not inducible with streptokinase.

These observations indicate that clot-selective coronary thrombolysis can be induced in patients with evolving myocardial infarction by means of tissue-type plasminogen activator, without concomitant induction of a systemic lytic state. Definition of its therapeutic benefit must await greater availability of the agent and the performance of appropriate clinical trials. (N Engl J Med 1984; 310:609-13.)

INITIATION of myocardial reperfusion by systemic activation of the fibrinolytic system was demonstrated more than 20 years ago,[1,2] and reperfusion by intracoronary administration of activators in pilot studies was reported as early as 1976.[3] Enthusiasm for the approach was kindled by the observations by Rentrop et al.,[4] who demonstrated that thrombolysis with intracoronary streptokinase restored angiographic patency to the occluded vessels supplying jeopardized myocardium in as many as 80 per cent of patients,[5] and by DeWood et al., who angiographically documented coronary thrombosis soon after the onset of symptoms in patients with evolving acute myocardial infarction.[6] Beneficial effects on global[7,8] and regional ventricular function,[9] perfusion,[10] and myocardial metabolic integrity[11-13] have been observed.

Activators such as streptokinase or urokinase have recently been given for the most part by the intracoronary route, in the hope of providing a high local concentration while minimizing the predisposition to systemic bleeding. Whether administration is intracoronary or intravenous,[14] streptokinase and urokinase activate the fibrinolytic system in the general circulation,[15] inducing what has been called a systemic lytic state.[16] The lytic state increases the risk of bleeding and is reflected by conversion of plasminogen to plasmin in the circulation, proteolytic depletion of circulating fibrinogen, accumulation of fibrinogen-degradation products, and consumption of circulating α_2-antiplasmin.

Since the potential benefit of coronary thrombolysis is markedly dependent on its prompt implementation[11,17] and safety — e.g., during surgery which is frequently needed soon after thrombolysis to correct high-grade residual stenosis — certain properties are desirable in an activator. These include lack of antigenicity, selectivity of action on a clot, avoidance of induction of a systemic lytic state, and a short biologic half-life so that hemostatic integrity can be restored promptly in patients requiring invasive procedures. Accordingly, we have been evaluating the potential utility of tissue-type plasminogen activator for coronary thrombolysis.[12]

Tissue-type plasminogen activator is a naturally occurring serine protease that activates the fibrinolytic system under physiologic conditions by converting plasminogen to plasmin. It does not bind avidly to circulating plasminogen ($K_M = 65\ \mu M$) but has a high affinity for fibrin ($K_d = 0.16\ \mu M$). Circulating plasminogen binds avidly to the tissue-type plasminogen activator–fibrin complex through the plasmino-

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gen-lysine binding sites ($K_M = 0.14\ \mu M$). Thus, plasminogen is not readily converted to plasmin in the circulation but is converted to plasmin at the fibrin surface of clot while still in association with fibrin. Any plasmin that escapes into the circulation is rapidly inactivated by α_2-antiplasmin (half-life, 100 msec).[16]

Recently, we isolated and purified human tissue-type plasminogen activator from a Bowes-melanoma–tissue-culture supernate[18] and administered it intravenously to dogs with coronary thrombi.[12] Thrombolysis was prompt and occurred without the marked depletion of circulating fibrinogen or plasminogen, accumulation of fibrinogen degradation products or consumption of circulating α_2-antiplasmin that would have been indicative of a systemic lytic state. The efficacy of tissue-type plasminogen activator in lysing clots in vivo has also been demonstrated in rabbits with jugular-vein thrombosis[19] or pulmonary emboli.[20]

This study was undertaken to determine whether administration of tissue-type plasminogen activator directly into the coronary arteries or intravenously could lyse coronary thrombi in patients with evolving acute myocardial infarction, and if so, whether lysis could be achieved without marked systemic fibrinogenolysis. The results indicated that tissue-type plasminogen activator could elicit coronary thrombolysis promptly without inducing a systemic lytic state.

Methods

Patients were studied only after their informed consent had been obtained and a human-studies committee had approved the investigation. This report summarizes our experience with all patients with imminent or established myocardial infarction who have been given the agent to date. The following criteria were applied for inclusion: a history of chest discomfort typical of myocardial ischemia persisting for at least 30 minutes, a 1.5-mm ST-segment elevation in at least two frontal-plane or two precordial electrocardiographic leads, feasibility of initiating thrombolytic therapy within 12 hours of the onset of symptoms, and age of less than 70 years. Exclusion criteria included Q waves indicative of completed or remote infarction, major illness other than coronary disease, contraindications to thrombolytic agents,[21] and concomitant anticoagulation. Infarction was documented by elevation of plasma enzymes in blood samples obtained before thrombolysis.

Catheterization of the right and left sides of the heart was performed conventionally according to the Sones or Judkins procedure. After right-heart hemodynamics and cardiac output were assessed by thermodilution and by introduction of a standby bipolar pacing electrode into the right ventricle, left-heart catheterization was performed with an angiographic pigtail catheter. Left ventriculograms were obtained (right anterior oblique projection) after left ventricular hemodynamics had been determined. Selective coronary arteriography was performed, with vessels assumed not to be involved by the infarct visualized first. When acute thrombotic occlusion was documented, tissue-type plasminogen activator was infused selectively or intravenously in the doses indicated in Table 1. Arteriography was repeated at selected intervals during the infusion and again after its completion. In two patients, infusions of tissue-type plasminogen activator were followed by infusion of streptokinase (Table 1).

Blood samples, treated with citrate to prevent coagulation, were obtained before, during, and after infusion of tissue-type plasminogen activator, cooled to 0 to 4°C, and centrifuged immediately. One aliquot of the decanted plasma, mixed with aprotinin (250 units per milliliter) to preclude proteolysis in vitro, was frozen and stored at −20°C until assays were performed for fibrinogen and fibrin (or fibrinogen) degradation products. Another aliquot was frozen without aprotinin, for assay of tissue-type plasminogen activator, plasminogen, and α_2-antiplasmin as previously described.[12]

Tissue-type plasminogen activator was extracted and purified from the melanoma cell-subculture as previously described[18,22] and in conformity with an Investigative New Drug application approved by the Food and Drug Administration. Streptokinase was purchased from Hoechst–Roussel (Streptase).

Except for administration of anticoagulants and activators of the fibrinolytic system, patients were treated conventionally. All patients given tissue-type plasminogen activator were then treated with heparin for a minimum of 24 hours at a dose of approximately 1000 units per hour. No bleeding complications were encountered before or after administration of thrombolytic therapy or heparin.

Table 1. Response to Tissue-Type Plasminogen Activator in Seven Men with Transmural Acute Myocardial Infarction.*

Patient No.	Age	Diagnosis	Locus of Occlusion	Duration of Symptoms †	Dose	Response	Remarks
	yr			*hr*	*IU*		
1	51	Anterolateral infarct	LAD	5	t-PA:1.3×10^6/60 min i.c.	Lysis in 50 min	Severe stenosis after lysis
2	62	Anterior infarct; CABG (1978)	SVBG to LAD	2.5	t-PA:1.0×10^6/45 min i.v. SK:250,000/30 min ‡	Lysis in 30 min, complete in 75	No complications
3	59	Anterior infarct; angina (3 yr)	LAD	2.5	t-PA:6×10^5/30 min i.v. t-PA:3×10^5/15 min i.c. SK:250,000/60 min i.c.	No lysis	Transient hypotension during SK infusion
4	57	Inferior infarct; angina (1 yr)	RCA	2	t-PA:6×10^5/30 min i.v. t-PA:3.6×10^5/18 min i.c.	Lysis in 30 min, complete in 33	Successful CABG 10 days after t-PA infusion
5	60	Inferior infarct; hypertension (5 yr)	RCA	2.25	t-PA:1.2×10^6/30 min i.v. t-PA:5.4×10^5/27 min i.c.	Lysis in 37 min, complete in 57	Infarction and CABG 5 days after t-PA infusion
6	68	Anterior infarct; hypertension (10 yr)	LAD	1.2	t-PA:1.4×10^6/35 min i.v.	Lysis in 22 min complete in 35	Reocclusion and reinfarction 18 days after t-PA infusion
7	63	Anterior infarct	Proximal LAD	3.3	t-PA:1.2×10^6/30 min i.v.	Lysis in 19 min	No complications

*CABG denotes coronary-artery bypass graft, i.c. intracoronary, i.v., intravenous, LAD left anterior descending coronary artery, RCA right coronary artery, SVBG saphenous-vein bypass graft, SK streptokinase, and t-PA tissue-type plasminogen activator.

†Before angiography.

‡When the small supply of activator available in this case was exhausted, streptokinase was given in a dose designed to be equipotent for thrombolysis, to permit comparison of effects of the two agents.

Results

As indicated in Table 1, seven patients with acute coronary thrombosis were given infusions of tissue-type plasminogen activator. Clot lysis was achieved in six of the seven. Representative coronary angiograms obtained in Patient 1, in whom thrombolysis was induced with tissue-type plasminogen activator administration initiated five hours after the onset of symptoms, demonstrated restoration of patency to the occluded vessel (Fig. 1). As was frequently the case, residual, high-grade stenosis was evident. Once an appropriate intravenous dose had been determined, lysis was successfully accomplished with intravenous infusions (Patients 4, 6, and 7) (Table 1). Since this study was undertaken to obtain initial experience, it was not possible to standardize the dose or route of administration a priori. Intracoronary administration of tissue-type plasminogen activator was used initially in some patients, intravenous followed by intracoronary administration was used in others, and streptokinase was administered after tissue-type plasminogen activator in the one patient whose occlusion proved refractory to thrombolysis and in another patient when the available supply of tissue-type plasminogen activator became exhausted after thrombolysis had been established. The one clot that was refractory to tissue-type plasminogen activator was refractory to streptokinase as well.

None of the patients had any complications related to the infusion of tissue-type plasminogen activator. Administration of tissue-type plasminogen activator did not induce a systemic lytic state (Fig. 2), in contrast to the usual case when streptokinase or urokinase is administered by intracoronary or intravenous routes in conventional doses.[21] Blood samples were obtained immediately before and after infusion of tissue-type plasminogen activator and immediately after infusion of streptokinase. Tissue-type plasminogen activator was assayed immunoradiometrically.[23] Fibrinogen-degradation products were assayed by hemagglutination inhibition,[24] fibrinolytic and activator activity by the fibrin plate assay,[24] and fibrinogen, plasminogen, and α_2-antiplasmin as previously described.[12] Tissue-type plasminogen activator concentrations averaged 25 ng per milliliter before infusion (range, 16 to 30) and 77 ng per milliliter after infusion (range, 38 to 118), with peak values during infusion averaging 481 ng per milliliter (range, 279 to 830). Fibrinolytic activity averaged 0.4 IU per milliliter (range, 0 to 1.3) before and 17 IU per milliliter after activator administration (range, 13 to 20). In the two patients given streptokinase after tissue-type plasminogen activator, fibrinolytic activity remained elevated, averaging 13.5 IU per milliliter, (range, 8 to 19). Absolute values for fibrinogen concentrations before infusion of activator (expressed as percentages of values in pooled plasma from normal subjects) ranged from 96 to 632 mg per deciliter and averaged 263 mg per deciliter. The findings that fibrinogen, plasminogen, and α_2-antiplasmin were not depleted and that fibrinogen-degradation products were not increased in response to infusion of tissue-type plasminogen activator reflect the lack of induction of a systemic lytic state. The values for these four features of the fibrinolytic system during infusion of the activator did not differ appreciably from those measured immediately afterward. In contrast to tissue-type plasminogen activator, streptokinase decreased fibrinogen, elevated fibrinogen-degradation products, decreased plasminogen, and diminished α_2-antiplasmin in 2 patients in this study and 17 patients with transmural myocardial infarction treated at Washington University with intracoronary streptokinase (250×10^6 to 750×10^6 IU) for clinical indications.[13] Results of other laboratory tests of hemostasis, including prothrombin time, activated partial thromboplastin time, rapidly clottable fibrinogen (Claus method), protamine-neutralized thrombin time, and platelet counts, remained normal during and after infusion of tissue-type plasminogen activator.

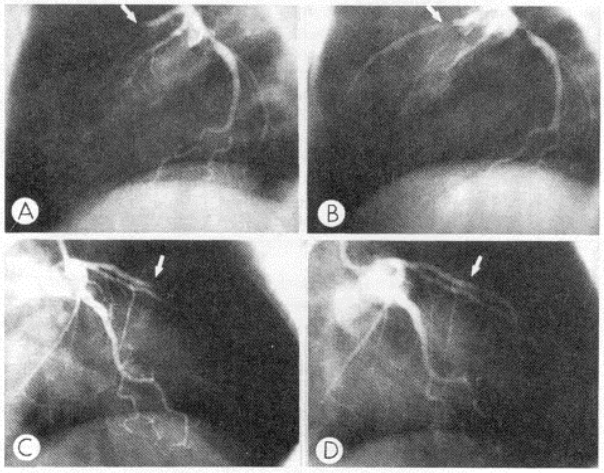

Figure 1. Left Anterior Descending Coronary Artery before (Panels A and C) and after (Panels B and D) Clot Thrombolysis by Infusion of Tissue-Type Plasminogen Activator in Patient 1.
Panels A and B show left anterior oblique projections, and Panels C and D right anterior oblique projections. Infusion restored patency despite residual high-grade stenosis (arrows).

This study was not performed to delineate the long-term effects of coronary thrombolysis induced with tissue-type plasminogen activator on myocardial performance, morbidity, or mortality after myocardial infarction. However, ventriculography performed before and after thrombolysis demonstrated either no change in apparent global or regional ventricular function, or an improvement (which was striking in two patients).

All patients studied survived the episode of acute infarction. Nevertheless, as anticipated in view of the severity of underlying disease, residual, high-grade stenosis was persistent after thrombolysis in four patients (Patients 1, 4, 5, and 6), reinfarction occurred in

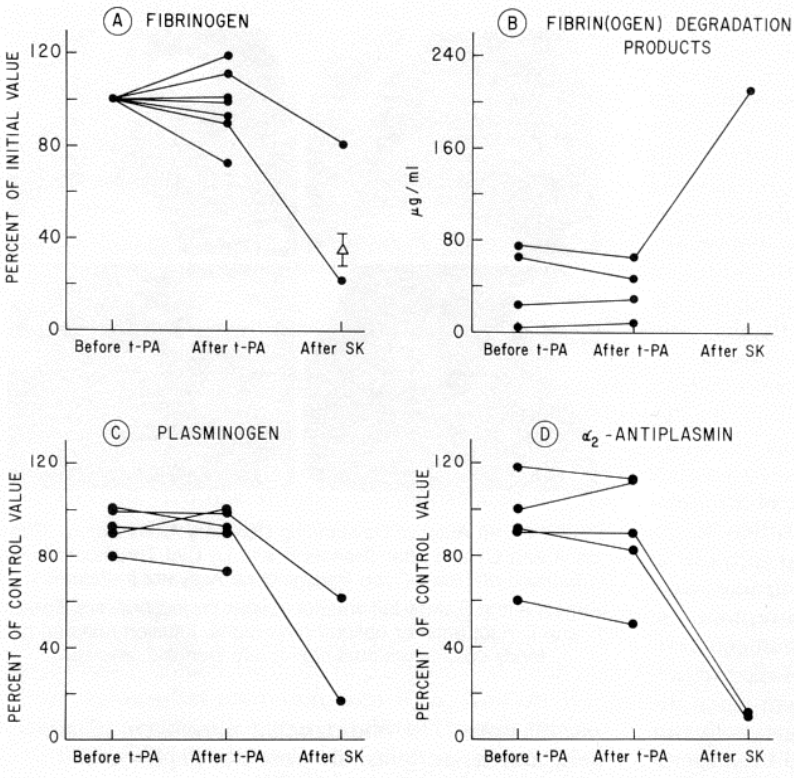

Figure 2. Effect of Tissue-Type Plasminogen Activator (t-PA) and Streptokinase (SK) on the Fibrinolytic Systems of the Patients.

For a description, see Results. The triangle (Panel A) denotes the mean ±S.E. of results in 17 other patients treated with intracoronary streptokinase alone.[13]

two, emergency coronary-artery surgery was required five days after thrombolysis in one, and elective coronary-artery surgery was performed 10 days after thrombolysis in another.

Discussion

The results obtained indicate that both intracoronary and intravenous tissue-type plasminogen activator can elicit coronary thrombolysis without inducing a systemic lytic state. The clot-selective properties of this agent make it an attractive candidate for coronary thrombolysis, since early intravenous administration of large doses of activator could increase the likelihood of early reperfusion.

Apparently beneficial effects of coronary thrombolysis include prompt reduction of chest pain, resolution of ST-segment abnormalities, and improved ventricular function.[8,15,25] However, although early reperfusion has salvaged myocardium in animals, it is not yet clear whether coronary thrombolysis confers long-term benefit on patients. Interpretation of results of clinical studies is confounded by several unavoidable limitations. On the one hand, lysis must be implemented early if it is to be effective.[11] On the other hand, treatment of patients with suspected but unconfirmed infarction may bias results in favor of the intervention.[15] The electrocardiogram may be misleading with respect to the ultimate extent of injury sustained, because of the altered sequence and time course of changes in response to reperfusion, and interpretation of plasma enzyme time–activity curves is clouded by possible effects of accelerated washout of enzyme. Improvement of global ventricular performance may reflect altered myocardial turgor or afterload, or changes of function in uncompromised regions, rather than direct effects on myocardial viability.

Nevertheless, findings from several studies suggest that coronary thrombolysis may abort or limit infarction, allow time for implementation of more definitive therapy in high-risk patients, and possibly provide long-term benefit.[26] Its ultimate value therapeutically is likely to depend in part on the extent to which complications such as systemic bleeding can be avoided and the safety with which high doses of activator can be promptly administered systemically in patients with suspected infarction.

The results of this study should not be interpreted too broadly. Our goal was to delineate properties of the activator pertinent to coronary thrombolysis. Definition of the therapeutic benefit, or lack thereof, of thrombolysis will undoubtedly require large-scale, randomized, prospective, blinded clinical trials. The small number of patients whom we studied does not permit accurate estimation of the frequency with which tissue-type plasminogen activator will lyse clots or delineation of characteristics of the dose–response curve. Most of our patients had occlusions of the left anterior descending coronary artery. Occlusions of the circumflex artery may not be equally responsive to tissue-type plasminogen activator. However, judging from results in laboratory animals,[11,12] the particular locus of an occlusion does not appear to be a critical determinant of its response to the activator. Since all our patients were men, we cannot be certain that the agent will be equally effective in women. Our patients were generally hospitalized at the time of onset of symptoms. Thus, the activator could be administered relatively early in their course. It is not possible to compare the relative efficacy of different activators unless they are used at equivalent doses in large numbers of patients with thrombosis of comparable duration. Nevertheless, the preliminary experience reported here suggests that tissue-type plasminogen activator is an effective and safe coronary thrombolytic agent.

Widespread use of tissue-type plasminogen activa-

tor or any activator requires a ready supply of active material. Isolation and purification of protein from tissue-culture supernatant fractions, as in this study, are not likely to meet the need for large-scale availability of preparations. Recent reports of successful cloning and expression of the human tissue-type plasminogen activator gene in *Escherichia coli*[27] suggest that large-scale production can be accomplished with recombinant-DNA technology. We have recently shown that the recombinant-DNA gene product has coronary thrombolytic activity in dogs comparable to that of the tissue-type plasminogen activator used in the present study.[28]

Since tissue-type plasminogen activator is a circulating human protein, the likelihood of induction of allergic reactions is low. Its short half-life in the circulation (approximately five minutes)[16] may be helpful for patients who require surgery or other invasive procedures soon after lysis has been achieved. The absence of induction of a systemic lytic state is likely to benefit hospitalized patients who have often had invasive procedures and in whom the risk of bleeding is appreciable. Thus, tissue-type plasminogen activator offers promise for clot-selective coronary thrombolysis in patients with evolving myocardial infarction.

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