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THERAPEUTIC FIBRINOLYSIS WITH TPA FAILED BECAUSE TPA'S MECHANISM OF ACTION WAS MISUNDERSTOOD

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ABSTRACT

Therapeutic fibrinolysis has used tissue plasminogen activator (tPA) since 1987 based on the belief that tPA was alone responsible for fibrinolysis. This was, however belied by clinical experience with tPA as well as experimental findings showing that fibrinolysis required both biological activators. In the clinic, it was never possible to show that tPA was more effective than the previous activator, streptokinase (SK), despite comparative mega trials in acute myocardial infarction (AMI) in which the only significant difference found was that tPA caused more intracranial hemorrhage (ICH) than SK. Eventually, this experience with tPA resulted in tPA as well as fibrinolysis being discredited and replaced by percutaneous coronary intervention (PCI) whenever possible. In ischemic stroke, fibrinolysis with tPA remains the standard. By contrast, the endogenous, physiological system induces fibrinolysis with both tPA and urokinase plasminogen activator (uPA). The native form of uPA is a proenzyme, prouPA, and tPA and prouPA have complementary modes of action making them synergistic in combination. This combination was once tested clinically in 101 AMI patients who were given a mini bolus of tPA (5 mg) followed by a modest infusion of prouPA. Compared with the results in the best of the tPA trials, this combination regimen almost doubled the infarct artery TMI-3 patency rate and reduced the mortality rate six-fold. The findings are a testimony to the therapeutic potential of the endogenous paradigm of fibrinolysis compared with current monotherapy.

Keywords: Nil

Introduction

The science philosopher, Thomas Kuhn, showed that "Science does not progress as a linear accumulation of new knowledge, but undergoes periodic revolutions called paradigm shifts." The paradigm from a well-established method is resisted, explaining why tPA treatment has persisted for so long despite not working well, and as a result, scientific progress is stalled. No change has occurred since 1987, despite a consensus that tPA is inadequate and its ICH complication risk is significant.

Instead, the treatment of choice has become PCI, which delays reperfusion significantly and precludes optimal salvage of myocardial and brain function. There are two plasminogen activators involved in the biological fibrinolysis, rather than only one. The other, uPA, the native form of which is a proenzyme, prouPA. tPA and prouPA have complementary modes of action are synergistic, which gives them an effect superior to that of either one alone.

The tPA clinical experience provides little support for its singular role in fibrinolysis. Even after comparative trials in 95,740 patients with AMI [1-3], it was not possible to establish that tPA was better than SK [4]. In these studies, the tPA dose used gave a plasma concentration about 1,000 fold higher than its physiological one, which was expected to be highly effective. Although the trial results contradicted the expectations and were not consistent with the tPA hypothesis, no explanation for the paradoxical findings was offered and tPA remained the unquestioned fibrinolytic of choice.

It is evident that a more effective and safer fibrinolytic regimen is needed.

Fibrinolysis requires both biological plasminogen activators is the obvious answer

Gene deletion studies

Gene knockout studies in mice showed that deleting the tPA gene had no measurable inhibiting effect on lysis of an intravascular clot and did not induce significant spontaneous fibrin deposition. By contrast, when the uPA gene was deleted, both inhibition of clot lysis and spontaneous fibrin deposition occurred. When both genes were deleted, a significantly stronger effect on both of these measures took place. The investigators concluded that both activators were required for a full fibrinolytic effect [5] but that uPA rather than tPA had the dominant effect [5, 6]. This finding reflects uPA having two functional states, single-chain proenzyme and two-chain enzyme, whereas tPA's single and two-chain forms have the same activities [8]. Although the gene deletion studies were published as much as 20 years ago in prominent journals, the findings were ignored.

Their complementary modes of action requires both for an optimal effect

Clot lysis studies in human plasma were consistent with the gene deletion studies. For example, the double gene knockout effect is explained by tPA and prouPA's complementary modes of action [17], so that both are needed for a complete affect and they have a synergistic fibrinolytic effect in combination

[18]. [18]. In a recent study, this synergistic effect was shown to be promoted almost two-fold when the activators were used sequentially, starting with tPA [19].

This is also how the endogenous system functions in which fibrinolysis is initiated by tPA when it is released from the vessel wall at the site a thrombus, and is continued by prouPA in the blood that is activated to tcuPA during lysis. Although some of these finding go back almost 30 years, fibrinolytic therapy with tPA alone continued without ever being put into question.

Clinical corroboration of the sequential combination effect

A sequential administration of the two activators was once tested in 101 patients with AMI. In the first 10 patients, a 10 mg bolus of tPA was given to initiate lysis, which turned out to be excessive and so only a 5 mg bolus (5% of the standard tPA dose) was given to the remaining 91 patients. This was followed by an infusion of prouPA, 40 mg/h for 90 minutes [34]. This combination induced a TIMI-3 patency at 24 h 82% of the 28 patients re-catheterized at that time, the overall mortality was 1%. This compares favorably with the best results in the tPA trials in which the TIMI-3 patency at 24h was 45% and the mortality was 6.3% [3].

Despite this high infarct artery patency and 6-fold reduction in mortality by this combination, the study had no effect on fibrinolytic therapy with tPA alone, and a second study was never done. This was because the pharmaceutical company developing prouPA was sold to Pharmacia which decided to discontinue its cardiovascular product line.

Endogenous fibrinolysis utilizes both activators

The endogenous fibrinolytic system functions efficiently with a tPA plasma concentration of only 10-12 ng/mL, much of which is in an inactive complex with its inhibitor, plasminogen activator inhibitor-1 (PAI-1) [20]. Nevertheless, lysis is sufficient to generate a concentration of 112-250 ng/mL of D-dimer in normal individuals. Since D-dimer represents ~60% of the fibrin monomer mass, this plasma concentration represents a steady state level of fibrin degradation of ~1 mg of fibrin. In patients with thromboembolism, D-dimer levels of ≥ 5,000 ng/mL are found, corresponding to of ≥ 25 mg of fibrin being degraded. In 15% of patients with AMI, the coronary thrombus responsible for the infarct was absent by the time the patient came to catheterization for primary PCI [22], representing the rate of endogenous fibrinolysis. This spontaneous TIMI-3 patency rate compares with one only three-fold greater at 24 h by tPA at a therapeutic concentration [3].

The efficacy of endogenous fibrinolysis cannot be explained by the small amount of tPA available alone, most of which is in an inactive inhibitor complex with PAI-1. Instead, it can be explained by the sequential effects of both activators in a synergistic combination, which is also consistent with findings from gene deletion findings [5,6] and clot lysis studies [15, 16]. Since prouPA is a proenzyme, it is not inhibited by plasma PAI-1, although there is little prouPA in plasma, most of it is carried by platelets [16] and monocytes [33], which gives it a longer plasma half-life.

The molecular function of biological fibrinolysis

The source of tPA in fibrinolysis is the vessel wall where it is stored and from where it is released at the site of an intravascular fibrin thrombus. The tPA then binds to the fibrin thrombus due to its exceptionally high affinity, a property which sets tPA apart from other plasma proteins [1]. The unbound tPA is rapidly cleared from the circulation (T1/2 ~5 min) or is inhibited by PAI-1, suggesting that free tPA is hazardous, which it is to hemostatic fibrin to which it can bind resulting in its disruption. This is the principle cause of bleeding during tPA therapy [33].

Therefore, tPA activates the first plasminogen on intact fibrin and prouPA and tcuPA the remaining two which are on partially degraded fibrin. Since tPA has only the single binding site at which fibrinolysis is initiated, it cannot activate the other two plasminogens, which are activated, in fact, by uPA.

Thus, tPA's specificity is to plasminogen on the D-domain of intact fibrin where the ternary complex is formed whereas prouPA's specificity is to plasminogen on the E-domain of partially degraded fibrin where the conformational change occurs. This fibrin-domain specificity was confirmed by a kinetic study with isolated fibrin fragments D and E. Plasminogen activation by tPA was promoted only by fibrin fragment-D and that by prouPA was promoted only by the fibrin fragment-E [12], confirming that their fibrinolytic effects are complementary and sequential. Both are required for a full effect to be also fibrin-specific so that bleeding complications are minimized or eliminated.

Conclusions

Prompt reperfusion of a thrombus blocked artery is essential for optimal salvage of heart or brain function and to achieve the lowest mortality. This is possible only with thrombolysis, which is a simple outpatient treatment. However, tPA alone is ineffective and hazardous due to the large doses required. PCI is too time-consuming. Fibrinolysis based on the natural fibrinolytic paradigm is highly effective and is safe due to the lower doses required by synergy. The efficacy of a sequential combination of a tPA bolus followed by a prouPA infusion was previously demonstrated in 101 AMI patients [18]. This doubled the infarct artery patency rate and reduced mortality 6-fold. A paradigm shift in the management of therapeutic fibrinolysis is long over-due.

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