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Coagulation and Fibrinolytic activity of tenecteplase and alteplase in acute ischaemic stroke

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Tables and figures:

Table 1. Key demographic and stroke characteristics of the 30 patients

Figure 1. The changes of coagulation and fibrinolytic variables in alteplase and tenecteplase treated stroke patients from baseline to 24 hours post thrombolysis.

Abstract

Background and purpose: We compared the fibrinolytic activity of tenecteplase and alteplase in acute ischemic stroke patients, and explored the association between hypofibrinogenaemia and intracerebral haemorrhage (ICH).

Methods: Venous blood samples from a sub-group of participants in the Alteplase-Tenecteplase Trial Evaluation for Stroke Thrombolysis (ATTEST) study were obtained at pre-treatment, 3-12 hours, and 24±3 hours post intravenous (IV) thrombolysis for analyses of plasminogen, Plasminogen Activator Inhibitor-1 (PAI-1), D-Dimer, Factor V, Fibrinogen, and Fibrin(ogen) Degradation Products (FDP), in addition to routine coagulation assays. Related sample Wilcoxon signed rank tests were used to test the within group changes, and independent Mann-Whitney tests for between group differences.

Results: 30 patients were included (Alteplase=14, tenecteplase=16), with similar baseline demographics. Compared to baseline, alteplase caused significant hypofibrinogenaemia (p=0.002), prolonged Prothrombin Time (PT) (p=0.011), hypoplasminogenaemia (p=0.001) and lower Factor V (p=0.002) at 3-12 hours after administration with persistent hypofibrinogenaemia at 24h (p=0.011), while only minor hypoplasminogenaemia (P=0.029) was seen in the tenecteplase group. Tenecteplase consumed less plasminogen (p<0.001) and fibrinogen (p=0.002) compared with alteplase.

Conclusion: In acute ischaemic stroke patients, alteplase 0.9mg/kg caused significant disruption of the fibrinolytic system while tenecteplase 0.25mg/kg did not, consistent with the trend towards lower ICH incidence with tenecteplase in the ATTEST study.

Clinical trial registration – URL: <http://www.clinicaltrials.gov> Unique identifier:

NCT01472926

Introduction

Intravenous (IV) thrombolysis with alteplase in acute ischaemic stroke (AIS) improves clinical outcome, but is associated with an absolute risk of fatal Intracerebral Haemorrhage (ICH) of around 2.7%, approximately 7-fold greater odds compared to placebo (OR [95%CI] 7.14 [3.98-12.79])¹. In two phase 2 trials in AIS^{2,3}, tenecteplase was associated with a trend towards fewer ICH complications.

As a sub-study of the Alteplase – Tenecteplase Trial Evaluation for Stroke Thrombolysis (ATTEST) study, we compared the effects of the two agents on coagulation and the fibrinolytic system, and explored potential associations with ICH.

Methods

The study protocol of ATTEST has been detailed elsewhere³. Eligible thrombolysis candidates within 4.5h of onset were randomised to receive a standard alteplase regime (0.9mg/kg) or 0.25mg/kg tenecteplase. This sub-study was initiated part-way through the main trial. All trial participants were approached after it commenced.

Venous blood samples were collected into citrate (final concentration 0.109 M, Greiner Bio-One, Austria) at baseline (pre-thrombolysis) (Time Point [TP] 1), 3-12 hours (TP2) and 24±3 hours (TP3) after the initiation of thrombolysis. Plasma was harvested by centrifugation immediately after sampling and stored at -80 °Celsius until analysis. We measured Prothrombin Time (PT), Activated Partial Thromboplastin Time (APTT), fibrinogen, Fibrin(ogen) Degradation Products (FDP), plasminogen, D-Dimer, Factor V (FV), PAI-1 activity, and prothrombin fragment 1+2 (F1+2) at three time points respectively (assay methods are detailed in supplementary material Table I.).

Statistical analysis

Baseline values were expressed as mean \pm standard deviation (SD), changes at TP 2 and TP3 as mean \pm SD percent change from baseline. We used related sample Wilcoxon signed rank test to examine the within-groups differences (TP2 vs TP1, TP3 vs TP1), using a Bonferroni correction to yield a significance level of $p < 0.025$. Between-group effects were explored with independent Mann-Whitney test.

Univariate binary logistic regression model was used to explore any association between the change of fibrinogen and ICH.

Results

Of 104 participants in the main ATTEST trial, 30 participated in this sub-study (alteplase=14, tenecteplase=16) (See supplementary material Figure I.). Key baseline characteristics were similar between groups (Table 1.).

Effects on coagulation and fibrinolysis (Figure 1.) (Supplementary material Table II.)

Alteplase was associated with prolongation of PT ($p=0.005$), reduced fibrinogen ($p=0.011$) and plasminogen ($p < 0.001$), elevated FDP ($p=0.002$) 24h post thrombolysis, a transient drop of Factor V ($p=0.002$) and increase of D-Dimer ($p=0.003$) at TP2. In contrast, tenecteplase resulted only in elevation of FDP ($p=0.009$), D-Dimer ($p=0.008$) up to 24h and transient reduction of plasminogen ($p=0.029$) at TP2.

Compared to tenecteplase, alteplase induced greater change of PT ($p=0.037$), fibrinogen ($p=0.002$), plasminogen ($p<0.001$) and Factor V ($p=0.002$) at TP2, with sustained differences in PT ($p=0.031$), fibrinogen ($p=0.011$) and plasminogen ($p=0.001$) at 24h.

Association between ICH and depletion of fibrinogen

Six patients had haemorrhage post thrombolysis (four with alteplase and two with tenecteplase), four classified as Haemorrhagic Infarction⁴ (HI)1, one as HI2, and one had a small Subarachnoid Haemorrhage (SAH). None was considered symptomatic using either ECASS 2⁴ or SITS-MOST criteria⁵.

Fibrinogen level dropped below 1g/L at TP2 in two patients, both of whom received alteplase (2/30, 14%). This low level persisted at TP3 in one, whose follow-up CT revealed HI2; the other patient's fibrinogen rose to 1.4g/L at TP3, who had SAH. Binary logistic regression found no association between ICH and the change of fibrinogen between TP2 and TP1 ($p=0.37$).

Discussion

Tenecteplase has 15-fold higher fibrin specificity than alteplase⁶. High fibrin affinity should translate into greater potency for thrombolysis, while preserving the integrity of systemic coagulation⁷. AIS trials suggest that tenecteplase may be associated with lower ICH risk with similar or superior recanalization compared to alteplase^{2,3}. In this sample, alteplase caused significant fibrinogen depletion and consumption of plasminogen, and degradation of Factor V, while tenecteplase did not.

Early degradation of fibrinogen is associated with the occurrence of ICH. Matosevic et al⁸ reported that within 6 hours post thrombolysis, a decrease of ≥ 2 g/L in fibrinogen level was

an independent predictor for bleeding of all kinds. Significant hypofibrinogaemia (a decrease of 2g/L or 50% from baseline) occurs in about one fifth of those receiving IV alteplase, whereas a tenecteplase dose escalation study⁹ showed no severe hypofibrinogaemia (fibrinogen <1g/dL) in any dose (0.1-0.5mg/kg) tested. Similarly, in our sample, tenecteplase treatment did not cause hypofibrinogaemia using either of these criteria. We could not replicate an association between hypofibrinogaemia and ICH, probably due to the small sample.

Limitations of our study include small sample size, variable time of sampling at TP2, and the low incidence of serious ICH (none having Parenchymal Haemorrhage, or symptomatic clinical deterioration attributable to haemorrhage). Nonetheless, we found significant changes in coagulation and fibrinolysis after IV alteplase consistent with the literature, and minimal disruption with tenecteplase, consistent with a potentially better safety profile for tenecteplase, with retained fibrinolytic efficacy.

Conclusion

In acute ischaemic stroke, tenecteplase caused significantly less disruption to the coagulation and fibrinolytic systems compared to alteplase. This finding was consistent with the trend towards reduced incidence of ICH observed in the ATTEST trial.

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Data safety monitoring committee: Prof. Kennedy R Lees, Dr. Mark Parsons, Dr. Christopher Weir;

External adjudicators: Prof. Michael Hill, Dr. Andrew Demchuk;

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Disclosures

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Table 1. Key demographic and stroke characteristics of the 30 patients

	Alteplase N=14	Tenecteplase N=16	P value
Age year (mean±SD)	70±12	69±15	0.95
Male (n, %)	10 (71%)	10 (63%)	0.71
OTT mins (mean±SD)	187±52	181±47	0.75
Baseline NIHSS (median, IQR)	10 (6-15)	11 (8-17)	0.58
Cardioembolic stroke (n, %)	8 (57%)	8 (50%)	0.7
Baseline vessel occlusion (n, %)	9 (64%)	8 (50%)	0.34
Large vessel occlusion (ICA, M1) (n, %)	6 (43%)	6 (38%)	0.7
Sampling time for TP2 hours (median, IQR, range)	5.3(4.8-10.1) [3.7-11.6]	4.4(3.9-11.8) [3-12.1]	0.27
Sampling time for TP3 hours (median, IQR, range)	23.8(23.1-24.6) [20.9-25.5]	23.9(23.5-24.6) [21.2-25.5]	0.62
Diurnal Sampling for TP2* (n, %)	1 (7%)	5 (31%)	0.3

ICA Internal Carotid Artery; M1 Middle Cerebral Artery M1 Segment; TP Time point; TP2, 2-12 hours post thrombolysis; TP3, 24±3 hours post thrombolysis; *Sampling time between 7 -9 am. Frequencies were compared using Chi-squared test and Fisher's test; Mean or median values were compared using independent T test and Mann-Whitney U test respectively.

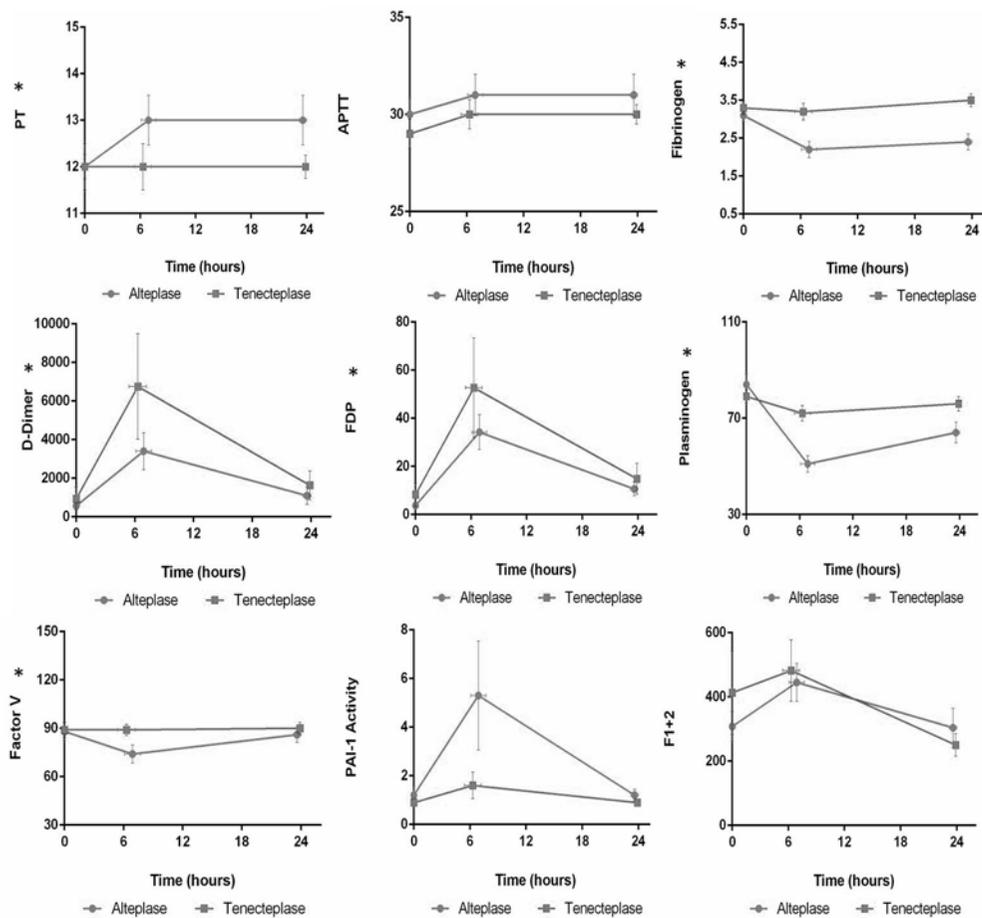


Figure 1. The changes of coagulation and fibrinolytic variables in alteplase and tenecteplase treated stroke patients from baseline to 24 hours post thrombolysis. PT, Prothrombin Time; APTT, Activated Partial Thromboplastin Time; FDP, Fibrin(ogen) Degradation Products; PAI-1, Plasminogen Activator Inhibitor-1; F1+2, Prothrombin Fragment 1+2. *Statistical significant difference within or between groups.