Tranexamic acid partially improves platelet function in patients treated with dual antiplatelet therapy


Background Although the impact of tranexamic acid on platelet function remains controversial, tranexamic acid is part of clinical algorithms for the management of platelet dysfunction. The goal of our prospective, observational study was to examine the effects of tranexamic acid on platelet function in patients treated with dual antiplatelet therapy compared to those who ceased antiplatelet therapy for at least 7 days.

Methods Forty patients scheduled for cardiac surgery were enrolled in this study. Group 1 consisted of 20 patients who ceased antiplatelet therapy with aspirin and clopidogrel at least 7 days before surgery. Group 2 consisted of 20 patients who were treated with aspirin and clopidogrel until the day before surgery. Using the Multiplate device (Dynabyte, Munich, Germany), multiple electrode aggregometry (MEA) was performed following platelet stimulation with thrombin receptor activating peptide-6 (TRAP-6), arachidonic acid or ADP on blood collected 20 min before and after application of 2 g tranexamic acid.

Results Compared with group 1, platelet aggregation was statistically significantly reduced in ASPTest and ADPTest in group 2, whereas there were no significant differences in the TRAPTest. In group 1, platelet aggregation did not differ significantly before and after tranexamic acid treatment. In contrast, in group 2, we observed a significant increase in arachidonic acid-induced [295 (280/470) arbitrary aggregation units x min [AU*min; median (25th/75th percentile) vs. 214 (83/409) AU*min, P = 0.01] and ADP-induced platelet aggregation [560 AU*min (400/760 AU*min) vs. 470 AU*min (282/550 AU*min), P = 0.013], whereas platelet aggregation following stimulation with TRAP-6 did not change significantly [980 (877/1009) AU*min, median (25th/75th percentile) after tranexamic acid vs. 867 (835/961) AU*min before tranexamic acid, P = 0.464].

Conclusion The results of this study indicate that tranexamic acid potentially corrects defects in arachidonic acid-induced and ADP-induced platelet aggregation imposed by dual antiplatelet therapy. However, platelet aggregation in response to arachidonic acid or ADP in the blood of patients who have not received aspirin and clopidogrel is unaffected by tranexamic acid. These results support the use of tranexamic acid to partially reverse platelet aggregation dysfunction due to antiplatelet therapy.

Published online 19 October 2010

Keywords: aspirin, clopidogrel, platelet aggregation inhibitors, platelet function test, tranexamic acid

Introduction Platelet dysfunction contributes to intraoperative and postoperative bleeding complications and might consequently increase perioperative mortality. With a prevalence of 3–5% in surgical patients collectively,1 perioperative impaired platelet function is a topic of high clinical relevance. Such platelet dysfunction is of multifactorial origin. Factors such as hypothermia,2 anaemia,3 interaction with colloids4 and exposure to foreign surfaces5 may lead to acquired platelet dysfunction. However, patients treated preoperatively with antiplatelet drugs for prophylaxis against ischaemic events are the predominant group of surgical patients suffering from platelet dysfunction.1 In this context, aspirin and clopidogrel are the most commonly used drugs.6

In cases of perioperative bleeding complications in patients with platelet dysfunction, transfusion of platelet concentrates represents a causal therapeutic approach. As controversial findings have been reported regarding whether platelet transfusions were associated with adverse outcome particularly in patients undergoing cardiac surgery or liver transplantation,7–9 pharmacological approaches to reverse the effects of antiplatelet therapy on platelet function are of high clinical relevance.

The lysine analogue tranexamic acid (Cyklokapron, MEDA Pharma GmbH, Bad Homburg, Germany) represents a facultative element of clinical therapy algorithms for the management of platelet dysfunction.10–12 Although the use of tranexamic acid as an antifibrinolytic drug was supported by a variety of clinical studies,13 up to now there is only a small number of publications with controversial findings which studied the effects of tranexamic acid on platelet function.14–16
Therefore, the aim of our prospective, observational study was to quantify effects of tranexamic acid on platelet aggregation in the blood of patients undergoing cardiac surgery and being treated with or without dual antiplatelet therapy. We assessed platelet aggregation in whole blood drawn before and after patients were given tranexamic acid using multiple electrode aggregometry (MEA) with the Multiplate point-of-care platelet function analyser (Dynabyte, Munich, Germany). MEA is a recently developed technique for platelet function testing in whole blood based on classical whole-blood impedance aggregometry.

Evidence is presented that the application of 2 g tranexamic acid corrects defects in arachidonic acid-induced and ADP-induced platelet aggregation in patients treated with dual antiplatelet therapy.

Patients and methods

Study population and intervention

This prospective, observational study complies with the Declaration of Helsinki and was approved by the local Ethics Committee which waived the necessity of patients to give written informed consent. A total of 40 adult patients scheduled for elective or urgent coronary artery bypass grafting (CABG) undergoing extracorporeal circulation were enrolled into this study. The study population consisted of two groups. Group 1 consisted of 20 consecutive patients who ceased antiplatelet therapy with aspirin and clopidogrel at least 7 days prior to surgery following local institutional standards for long-term scheduled elective cardiac surgery patients. Group 2 consisted of 20 consecutive patients who were urgently scheduled for cardiac surgery and who continued antiplatelet therapy with aspirin and clopidogrel until the day before surgery. According to our institutional standards, all patients received a cumulative dosage of 6 g tranexamic acid to prevent hyperfibrinolysis: a bolus of 2 g before skin incision, a bolus of 2 g into the priming volume of the extracorporeal circulation and a bolus of 2 g after aortic declamping. In order to assess the effect of tranexamic acid on platelet function specially, independent of factors such as hypothermia or any surgical stressors, the tests were done on blood collected before and after the first tranexamic acid dose, before incision.

Blood was drawn from a central venous line. The first 10 ml blood was discarded; then blood was collected into a 2 ml heparin-anticoagulated and calcium-balanced tube (Bloodgas-Monovette, Sarstedt AG).

Platelet function analysis

Platelet function analysis was performed using MEA, a whole-blood impedance aggregometer (Multiplate; Dynabyte). Analysis is based on aggregation of activated platelets onto metal sensor wires which increases the electrical impedance between the wires. For each measurement, 300 µl of preheated saline (37°C) and 300 µl of heparin-anticoagulated whole blood were placed into the test cell and the sample was stirred using a teflon-coated electromagnetic stirrer (800 revolutions min⁻¹) over a 3-min incubation period. Platelet aggregation was initiated with thrombin receptor activating peptide-6 (TRAPtest, TRAP-6, 32 µmol l⁻¹), arachidonic acid (ASPItest, AA, 0.5 mmol l⁻¹) or adenosine diphosphate (ADPtest, ADP, 6.4 µmol l⁻¹), using commercially available reagents (Dynabyte). Increased impedance due to attachment of platelets to the electrodes was continuously and separately measured for each sensor unit over 6 min. Data were transformed into arbitrary aggregation units and plotted against time. For internal quality control, two curves are plotted in each test. In the absence of any methodical errors, the two curves run parallel. Aggregation measured by MEA was quantified as the area under the aggregation curve [AUC, arbitrary aggregation units × min (AU*min)]. Reference ranges for healthy individuals obtained from the manufacturer were 868–1473 AU*min for the TRAPtest, 505–1086 AU*min for the ASPItest and 607–963 AU*min for the ADPtest. Values measured lower than normal indicate inhibition of platelet function in response to the specific agonist.

Statistics

Sample size analysis (expected difference of means for ASPItest 100 AU*min, expected SD 150 AU*min, desired power 0.8, and \( P < 0.05 \)) was based on previous experience of our study group with DDAVP and revealed a required sample size of at least \( n = 20 \) in group 2 in order to detect statistically significant differences in platelet aggregation before and after treatment.

The \( t \)-tests were used to detect differences in the two groups’ demographic data. A Mann–Whitney rank–sum test was used to analyse differences in conventional coagulation tests and baseline measurements in MEA between groups 1 and 2. Wilcoxon’s signed rank test was used to detect differences in MEA analyses before and after administration of tranexamic acid. Depending on the distribution on the data (Kolmogorov–Smirnov test), values were expressed as mean ± SD or median (with 25th/75th percentiles). The level of statistical significance was set at \( P < 0.05 \). Statistical analysis was performed using SigmaStat (version 3.5, Systat Software GmbH, Erkrath, Germany) and SigmaPlot (version 11, Systat Software GmbH, Erkrath, Germany) software.

Results

There were no differences in basic demographic data between the two groups (Table 1). Preoperatively performed standard laboratory coagulation analyses did not differ between the two groups (Table 2). There were no group differences in preoperative serum creatinine [1 (0.8/1.2) mg dl⁻¹ in group 1 vs. 1 (0.8/1.2) mg dl⁻¹ in group
Tranexamic acid improves platelet aggregation

Table 1 Demographic data of the two study groups

<table>
<thead>
<tr>
<th></th>
<th>Group 1 (n = 20)</th>
<th>Group 2 (n = 20)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex (male/female)</td>
<td>15/5</td>
<td>17/8</td>
<td>0.858</td>
</tr>
<tr>
<td>Age (years)</td>
<td>66 ± 8</td>
<td>66 ± 10</td>
<td>0.179</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>27 ± 4.1</td>
<td>29 ± 4</td>
<td>0.013</td>
</tr>
</tbody>
</table>

Age and BMI are given as mean ± SD.

2 (P = 0.942)]. Overall, 29 patients (n = 13 in group 1, n = 16 in group 2) underwent CABG and 11 patients (n = 7 in group 1, n = 4 in group 2) underwent combined procedures (valve surgery and CABG). The duration of extracorporeal circulation was 112 ± 49 min in group 1 and 99 ± 51 min in group 2 (P = 0.412). None of the patients underwent re-do procedures and none of the patients required postoperative surgical re-exploration.

Comparing the baseline parameters (before tranexamic acid) of each group, platelet aggregation in the TRAPtest revealed no significant group differences (P = 0.207), whereas there were significant group differences in platelet aggregation following stimulation with arachidonic acid (ASPItest, P < 0.001) and ADP (ASPItest, P < 0.001).

In those patients who ceased dual antiplatelet therapy for at least 7 days, we did not observe any significant change in platelet aggregation in neither the TRAPtest (P = 0.261), or in the ASPItest (P = 0.294), or in the ADPtest (P = 0.167) after the application of tranexamic acid (group 1, Fig. 1). In contrast, in those patients who did not cease dual antiplatelet therapy, we observed a significant increase in arachidonic acid-induced [295 (280/470) AU*min vs. 214 (83/409) AU*min, P = 0.01] and ADP-induced platelet aggregation [560 AU*min (400/760 AU*min) vs. 470 AU*min (282/550 AU*min), P = 0.013], whereas platelet aggregation following stimulation with thrombin receptor activating peptide-6 did not change significantly [980 (877/1009) AU*min, median (25th/75th percentile) after tranexamic acid vs. 867 (835/961) AU*min before tranexamic acid, P = 0.464].

Discussion

The main result of the present study was that application of the lysine analogue tranexamic acid resulted in a statistically significant increase in both arachidonic acid-induced and ADP-induced platelet aggregation selectively in those patients who did not cease preoperative dual antiplatelet therapy with aspirin and clopidogrel.

In the present study, platelet function was analysed using the MEA device performing whole-blood analyses. In fact, light transmission aggregometry (LTA) using platelet-rich plasma is considered to be the gold standard for assessing the platelet response to agonists such as arachidonic acid or ADP. However, LTA measurements require a specialised laboratory, are time consuming and weakly standardised. Furthermore, the logistical demands of LTA make it difficult to use in daily clinical practice. In contrast, the MEA is based on impedance aggregometry which was first described by Cardinal and Flower. MEA does not require a specialised laboratory and is useful for point-of-care analysis due to its use of a single-use test cell, integrated computer analysis and documentation and electronic pipetting. Its high specificity and sensitivity in platelet function analyses have already been demonstrated in several studies, also in comparison to LTA.

Corresponding to the aspirin-induced inhibition of the cyclooxygenase pathway and the clopidogrel-induced blockade of the ADP receptor pathway, arachidonic acid-induced and ADP-induced platelet aggregation in group 2 were below the lower reference ranges for normal aggregation before the application of tranexamic acid (Fig. 2). The relatively large 25th/75th interval in the ASPItest and ADPtest potentially indicates an interindividual variability in drug response to aspirin and clopidogrel. However, in none of the patients, ex-vivo aggregation was within normal reference ranges, indicating that none of the patients was ‘resistant’ to aspirin and/or clopidogrel. In contrast, as neither the arachidonic acid-induced pathway of platelet aggregation via cyclooxygenase 1 nor the ADP receptor were affected by any pharmacologic therapy in group 1, platelet aggregation in the corresponding tests (ASPItest, ADPtest; Fig. 1) was within normal ranges and was significantly higher as compared with group 2. Ex-vivo platelet aggregation following the TRAPtest was within normal range in both groups because neither the platelets thrombin receptor nor the glycoprotein IIb/IIIa receptor was affected by either medication.

Arachidonic acid-induced and ADP-induced platelet aggregation in MEA were thought to be dependent on the platelet count, haematocrit and general functional integrity of platelets. As we did not detect any differences in either conventional coagulation parameters, haematocrit or platelet count (Table 2) or in thrombin receptor-induced platelet aggregation in the TRAPtest at baseline in the two groups, our results indicate tranexamic acid treatment effects. As platelet aggregation in the ASPItest and the ADPtest increased equally, we suspected that there was a ‘direct’ effect of

Table 2 Preoperative analysis of conventional coagulation parameters

<table>
<thead>
<tr>
<th></th>
<th>Group 1 (n = 20)</th>
<th>Group 2 (n = 20)</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>Platelet count (nL⁻¹)</td>
<td>230 (187/262)</td>
<td>249 (191/278)</td>
<td>0.513</td>
</tr>
<tr>
<td>Haematocrit (%)</td>
<td>40 (38/43)</td>
<td>40 (38/44)</td>
<td>0.871</td>
</tr>
<tr>
<td>aPTT (s)</td>
<td>38 (35/40)</td>
<td>38 (35/38)</td>
<td>0.114</td>
</tr>
<tr>
<td>INR</td>
<td>1.3 (1.2/1.5)</td>
<td>1.4 (1.3/1.5)</td>
<td>0.401</td>
</tr>
<tr>
<td>Fibrinogen (mg/dl)</td>
<td>341 (284/395)</td>
<td>363 (307/410)</td>
<td>0.371</td>
</tr>
</tbody>
</table>

INR, international normalised ratio; PTT, partial thromboplastin time. Values are given as median (25th/75th percentiles).
tranexamic acid on receptor-mediated platelet aggregation. Two other studies including patients suffering from chronic renal failure have investigated the influence of tranexamic acid on platelet function. Performing in-vitro closure time measurements with the Platelet Function Analyzer PFA-100 (Siemens Healthcare Diagnostics, Eschborn, Germany) and investigating the skin bleeding time, they assumed that tranexamic acid treatment

![Figure 1](image1.png)

Platelet aggregation expressed in arbitrary units × min (AU^* min) before and after tranexamic acid treatment in patients who ceased dual antiplatelet therapy for at least 7 days prior to surgery; box plots represent 25th/75th percentiles and median (group 1, n = 20); horizontal dashed lines indicate the lower reference values of each test for normal platelet aggregation obtained by the manufacturer. Platelet aggregation was induced with thrombin receptor activating peptide-6 in the TRAPtest, arachidonic acid in the ASPtest and ADP in the ADPtest.

![Figure 2](image2.png)

Platelet aggregation expressed in arbitrary units × min (AU^* min) before and after tranexamic acid treatment in patients who did not cease dual antiplatelet therapy with aspirin and clopidogrel before surgery; box plots represent 25th/75th percentiles and median; asterisk (*) indicates P < 0.05 (group 2, n = 20); horizontal dashed lines indicate the lower reference values of each test for normal platelet aggregation obtained by the manufacturer. Platelet aggregation was induced with thrombin receptor activating peptide-6 in the TRAPtest, arachidonic acid in the ASPtest and ADP in the ADPtest.
resulted in a significant improvement in platelet function.\textsuperscript{25,26} In that case, how can the observed improvement in platelet function be explained? Both platelet adhesion to the subendothelial matrix and platelet aggregation are mediated mainly via glycoprotein Ib and glycoprotein \( \text{IIb/IIIa} \) receptors.

The plasma proteinase plasmin is known to multifactorially affect platelet function. It was shown to induce platelet activation by stimulating the protease-activating receptor 4 (PAR4).\textsuperscript{27} Plasmin-induced platelet activation was described to be similar to platelet activation due to thrombin.\textsuperscript{10} Furthermore, plasmin was shown to activate the complement cascade by generating activated complement factors C3a and C5a,\textsuperscript{28} which are known to induce platelet dysfunction and contribute to perioperative haemorrhage.\textsuperscript{29–31} Finally, plasmin was shown to induce a proteolytic degradation and redistribution of platelet glycoprotein Ib and \( \text{IIb/IIIa} \) receptors and thereby reduce platelet adhesion and aggregation.\textsuperscript{32,33} In this context, Lindvall et al.\textsuperscript{34} described an improvement in platelet function following the substitution of aprotinin. This corroborates the hypothesis that the main effect of influencing platelets function by the use of antifibrinolytics may be due to a reduction of plasmin-induced platelet inhibition.

Hence, tranexamic acid reduces the plasma concentration of plasmin by blocking the lysine-binding sites of plasminogen, thereby preventing the binding of plasminogen to fibrin and the consecutive conversion of plasminogen to plasmin.\textsuperscript{35} Thus, tranexamic acid may preserve platelet function by mitigating the above-mentioned multifactorial plasmin-induced platelet dysfunction.

The present study did not assess either the concentration of platelet glycoprotein Ib or \( \text{IIb/IIIa} \) receptors nor the plasma level of fibrinogen, von Willebrand factor, plasmin or plasminogen. Therefore, final conclusions concerning the mechanism of action of tranexamic acid in increasing arachidonic acid-induced and ADP-induced platelet aggregation partially in patients treated with dual antiplatelet therapy could not be drawn.

There were some limitations to this study. First, the study population was small and observational. Study results may be prone to alpha and beta errors, especially as the power analysis was based on hypothetical variables. Second, we analysed only one time point after tranexamic acid administration, so no information about duration of the effect of tranexamic acid on platelet function could possibly be derived from the data. Third, platelet function in response to tranexamic acid was assessed only by MEA and not using the gold standard for platelet function testing, the LTA. This method might have provided different results.

Despite these limitations, this study may be of clinical relevance. Although ex-vivo platelet function did not return to normal values after the substitution of tranexamic acid, this study leads to the assumption, that tranexamic acid may represent a pharmacological option to partially increase platelet aggregation in patients treated with dual antiplatelet therapy who come for cardiac or noncardiac surgery. As the design of the present study did not allow us to draw any conclusions concerning causal relationships between tranexamic acid and platelet function due to methodical limitations, randomised and placebo-controlled studies on larger study populations that also include healthy volunteers without any pharmacological therapy are needed to confirm our findings and to evaluate the potentially dose-dependent applicability of tranexamic acid to partially reverse the effects of dual antiplatelet therapy.

Acknowledgements

The study was performed with departmental funding from the Clinic for Anaesthesiology, Intensive Care Medicine and Pain Therapy, J.-W. Goethe University Hospital, Frankfurt, Germany. K.G. received speaking honoraria from Dynabyte, Munich, Germany.

References


European Journal of Anaesthesiology 2011, Vol 28 No 1

Copyright © European Society of Anaestheiology. Unauthorized reproduction of this article is prohibited.


