Transfusion Medicine and Hemotherapy

Letter to the Editors · Brief an die Herausgeber

Transfus Med Hemother 2008;35:324–326
DOI: 10.1159/000143229

Published online: July 21, 2008

Concerning Hänecke P, Klouche M: Thrombelastography Today: Practicability and Analytical Power. Transfus Med Hemother 2007;34:421–428

Thomas Ebinger

Pentapharm GmbH, München, Germany

The article 'Thrombelastography Today: Practicability and Analytical Power' by Hänecke and Klouche [1] is interesting in the respect that it warns users to follow strict rules and avoid errors to ensure result quality.

The article however requires some comments from the system manufacturer side: Hänecke and Klouche are not consistent about instrument name and test nomenclature.

In 2003, due to the registration of the trademark TEG® by a US-based company, Pentapharm took the decision to differentiate its largely shock insensitive method from the pendulum-based, shock-sensitive, earlier thrombelastographic methods. The Pentapharm method was then renamed to thromboelatometry, the instrument name changed to ROTEM® and all test names to XXTEM (e.g. INTEM). All reagents were trademarked xx-TEM® (e.g. in-TEM®). To avoid problems with the rights owner of the TEG trademark, authors should respect the trademark and not use 'TEG' in connection with the Pentapharm product.

Pentapharm welcomes the scientific approach for evaluation of the ROTEM® system with the emphasis on general operation conditions and site specifications in a laboratory environment. Pentapharm would however have wished this to happen using a more recent instrument model following Pentapharm instructions for use.

Pentapharm GmbH started refining the original instrument in 2002 with ROTEG 05; in 2004 the ROTEM® Gamma and in 2007 the ROTEM® *delta* as state of the art models were introduced. The authors did all their experiments using a 1999 ROTEG 04 model by the original inventor of the system.

Most of their points of critic have been known for a long time and have been addressed and solved since Pentapharm started refining the original instrument.

Practicability of Rotational Thrombelastography

In table 1, the authors show the influence of the ambient temperature fluctuation on the test results (especially on temperatures lower than a standard room temperature of 21 °C).

Many instruments must be operated in defined environmental conditions to ensure consistent results. Pentapharm discourages use of the system near an open window or an air conditioning outlet (user manual chapter 3.2). Furthermore users are advised to warm blood samples for approximately 10 min at 37 °C (e.g. in the sample pre-warming station of ROTEM® Gamma or ROTEM® *delta*) if the instrument is not used at the point of care (POC) and the sample cannot be tested immediately after sampling (user manual chapter 6.2.2).

In table 5, the precision of the automated pipette is tested at different volume settings: 10 µl, 20 µl and 300 µl; the results are found unsatisfactory. The minimum volume used in ROTEM® pipetting is 20 µl. Precision testing for 10 µl is therefore not necessary.

Since 2002, all Pentapharm made instruments are delivered with a calibrated and certified eLine pipette. Regulations concerning calibration oblige laboratories to check calibration of their pipettes in regular intervals. The user manual states the procedure and indicates acceptable errors of the pipette at various volumes (e.g. 1.0% at 20 µl). It also indicates that pipettes out of these ranges need to be sent to the manufacturer for maintenance and recalibration (ISO 8655). A new quality certificate will be supplied upon each calibration for the user's documentation (user manual chapter 14.10).

Hänecke and Klouche [1] state that extremely prolonged coagulation times in patients with coagulation disorders abrogate the potential saving of time. We, as many POC users, question this argument. ROTEM® is mainly used for detection of bleeding tendency and/or the case thereof in a perioperative setting. At a POC setting, the graphical result develop-

KARGER

Fax +49 761 4 52 07 14
E-mail Information@Karger.de
www.karger.com

Accessible online at:
www.karger.com/tmh

Dr. Thomas Ebinger
Pentapharm GmbH
Stahlgruberring 12, 81829 München, Germany

ment can be monitored dynamically. Any delay in clot formation in a patient sample can be interpreted as 'impaired coagulation'. The combination of 4 of the 5 ROTEM® tests (INTEM, EXTEM, FIBTEM, APTEM, HEPTEM) can lead to rapid targeted treatment [2–5].
Typical long clotting times (CT) are seen in heparinized patients using the intrinsic activator (INTEM). CT can be used to judge heparinization. To detect underlying pathologies, HEPTEM containing heparinase (or to some extent EXTEM) may be used. EXTEM-based ROTEM® tests also contain a heparin-neutralizing agent. Thus, the combination of e.g. an EXTEM and an INTEM test may be used to confirm or rule out accidental heparin administration.
Direct evaluation of the patient's coagulation status with ROTEM® at the POC is considered very rapid as compared to the return of laboratory results by many authors [4, 6–10].

Principle of Rotational Thrombelastography

Hänecke and Klouche [1] state that heparin and other drugs affect results. This is also true for many other tests in the clinical laboratory, and not a problem per se. It is always necessary to know possible limitations of a test. Respective warnings can be found in the instrument operator manual and the reagent insert sheets. Moreover, the heparin interference in certain tests is used to judge heparinization, and the protamin interference can be used to discriminate residual heparin effect from protamin overdosage [11].
Hänecke and Klouche [1] also warn for the unknown hemostatic potential of transfused blood products and their influence on the fibrinolytic system. They advise to take great care to interpret whole blood thromboelastography data from patients receiving transfusions. There we cannot agree. Today most authors agree that thromboelastometry is an excellent tool to monitor the effect of transfusions or of the component substitution therapy with blood components [2, 3, 12].
As thromboelastometry and thromboelastography visually show fulminant fibrinolysis, it is actually an excellent method to monitor the hemostatic state of hyperfibrinolytic patients after receiving treatment [4, 13].

Analytical Power of Rotational Thrombelastography

Hänecke and Klouche [1] claim that it is not possible to validate ROTEM® tests because it is impossible to provide a description of the incorrectness of the system for the following reasons:
i) there is no standardized control material available,
ii) there is no defined reference method,
iii) there is no definition of the target values to be expected.
However:
i) Two levels of standardized controls are available from Pentapharm: ROTROL N and ROTROL P. These control materials are plasma-based. Whole blood cannot be stabilized as a control material. This situation is not much different from blood gas analysis where even more artificial control material is accepted for the same reason: a whole blood control cannot be produced.
ii) In coagulation, definition of a reference method is generally much more difficult than in clinical chemistry, due to the complex processes measured. Even for tests as 'old' as prothrombin time and activated partial thromboplastin time, reference methods are still debated or not available. As thromboelastometric tests rely on whole blood, again the preparation of an international standard – the usual procedure in cases where a true reference method is not feasible – is not possible. Work towards standardization is ongoing. ROTEM® and the older, pendulum-based thrombelastographic methods correlate well [14, 15]. Correlation studies between e.g. ROTEM® FIBTEM MCF and Clauss fibrinogen are also available [10, 15].
iii) Reference ranges for each of the ROTEM® tests have been established in a multicentric study [16]. Tentative guidelines for trauma patients, heart surgery, liver transplant, and others have been published recently or publication is under way [2, 3, 17–19].

Quality Assessment

When assessing the performance of the system (table 3 and 4), Hänecke and Klouche [1] did not use the recommended ROTEM® reagents (star-TEM®, in-TEM®, ex-TEM®, fib-TEM®, ap-TEM® and hep-TEM®) but their own preparations based on Dade Behring Innovin® as extrinsic activator and Reopro® (Abciximab) as platelet inhibitor.
As sample type, the authors used not only the recommended citrated whole blood (user manual chapter 6.2) but also freshly prepared plasma. The obtained precision data for citrated whole blood were close to the ranges published by Pentapharm, despite the non-Pentapharm reagents.
For the freshly prepared plasma CVs obtained were high and particularly elevated for the clot formation time and the maximum clot firmness. This may be due to artifacts because of partial activation of the few remaining platelets during plasma preparation. As fresh plasma preparations are difficult to handle for thromboelastometry, Pentapharm does not recommend plasma as sample material!
The plasma based ROTROL N and ROTROL P control materials supplied by Pentapharm do however give reproducible results. There, the platelets are completely inactivated during the lyophilization process and the remaining fibrin clot is very reproducible. The conclusion of Hänecke and Klouche [1] that the use of the lyophilized ROTROL control material is not suitable according to their results with fresh plasma is not legitimate.

Quality Assurance

Hänecke and Klouche [1] are concerned about the lack of a quality assurance setup for ROTEM®.
Both UK-based NEQAS [20] and Germany-based Instand offered a first thromboelastometry/thromboelastography survey in 2007. Both schemes included 2 levels of lyophilized plasma and were designed for thromboelastometry (ROTEM®) and classical thromboelastography (e.g. TEG). The NEQAS results of the scheme were presented at the 2007 ISTH Congress in Geneva. In the USA, schemes from CAP and the American Proficiency Institute are available for thromboelastography.
For internal quality assurance, the Pentapharm control materials come with target values and ranges for each of the main ROTEM® parameters. In addition, a table with typical CVs for both citrated blood and lyophilized plasma for the various ROTEM® parameters is supplied by Pentapharm for customer orientation.
We completely agree with the authors that more still can be done to implement quality assurance measures at the customer site. But again, this is true for most of the POC methods and is primarily the responsibility of the hospitals. Industry can only help here by providing adequate means and information.

Instructive Case

In the instructive case, of Hänecke and Klouche [1] wanted to know if aspirin could have caused the bleeding in this patient with pronounced thrombocytosis.

The ROTEM® EXTEM test shows that the secondary hemostasis (as measured by ROTEM®) is not impaired. Therefore, if the patient continues bleeding (and surgical bleeding is excluded), additional testing of the primary hemostasis is required (e.g. Born aggregometry, Multiplate etc.). Current ROTEM® tests are not sensitive to impairment of primary hemostasis (user manual chapter 6.3 and 7.2). Questions concerning anti-platelet therapy can therefore not be answered by the ROTEM®.
From the TEMogram, the authors conclude that at 1.3×10^6 platelets/µl the FIBTEM amplitude may be false high and that the fib-TEM reagent (containing Ca^{2+} and cytochalasin D) may have failed to block all the platelets. This appears plausible and may require further evaluation. As very high platelet count in combination with bleeding is very rare in the operating room, few reports are available on this phenomenon. With platelet counts in the normal range, the fib-TEM reagent is able to block the available platelets. Pentapharm has received reports from patients with higher platelet counts where the fib-TEM reagent had no difficulty completely inhibiting platelet counts of approximately 650,000/µl (personal communication Dr. DiFlorio, Napels, Italy).
Recently a study of this effect has been initiated by Pentapharm.
We completely agree with the authors that patient treatment should never be based on single test results but as many results as available together with the patient history. In addition, the clinical picture should be considered at all times for rational and targeted treatment.

References

1 Hänecke P, Klouche M: Thrombelastography today: practicability and analytical power. Transfus Med Hemother 2007;34:421–428.
2 Anderson L, Quasim I, Steven R, Soutar M, Macfie A, Korte W: An audit of red cell and blood product use after institution of thromboelastography in a cardiac intensive care unit. Tranfus Med 2006;16: 31–39.
3 Innerhofer P: Perioperatives Gerinnungsmanagement. Hämostaseologie 2006;26(suppl 1):S3–S14.
4 Vorweg M, Doehn M, Kolde H-J: Rapid diagnosis and differentiation of acute haemostasis disorders with whole blood global assays Eur Clin Lab 2002; 8:12–15.
5 Vorweg M, Hartmann B, Knüttgen D, Jahn MC, Doehn M: Management of fulminant fibrinolysis during abdominal aortic surgery. J Cardiothorac Vasc Anesth 2001;15:764–767.
6 Calatzis AN, Fritsche P., Kling M., Calatzis AL, Mielke L, Stemberger A: A new technique for fast and specific coagulation monitoring Eur Surg Res 1996;28(suppl 1):89.
7 Calatzis AN, Kling M, Calatzis AL, Mielke L, Hipp R, Stemberger A: Fast and specific coagulation monitoring through modified thrombelastography J Cardiothor Vasc Anesth 1994;8:144–149.
8 Koster A, Fischer T, Kukucka M, Kuppe H: A multichannel TEG based synoptic approach for rapid detection of postoperative coagulation disorders in pediatric patients following cardiac surgery ASA 2000;abstract A-1312.
9 Kozek-Langenecker S: Management of massive operative blood loss Minerva Anestesiol 2007;73: 401–415.
10 Rugeri L, Levrat A, David JS, Delecroix E, Floccard B, Gros A, Allaouchiche B, Négrier C: Diagnosis of early coagulation abnormalities in trauma patients by rotation thrombelastography. J Thromb Haemost 2007;5:289–295.
11 Mittermayer M, Margreiter J, Velik-Salchner C, Klingler A, Streif W, Fries D, Innerhofer P: Effects of protamine and heparin can be detected and easily differentiated by modified thromboelastography (ROTEM®): an in vitro study Br J Anaesth 2005;95:310–316.
12 Innerhofer P, Streif W, Kühbacher G, Fries D: Monitoring of perioperative dilutional coagulopathy using the ROTEM® analyzer: basic principles and clinical examples. Transfus Med Hemother 2004;31: 244.
13 Calatzis AL, Calatzis AN, Kling M, Mielke L, Hipp R, Stemberger A: Fast detection of fibrinolysis by two parallel thromboelastographic measurements: APTEG and EXTEG Ann Hematol 1996;73(suppl I):P138.
14 Calatzis An, Calatzis AM, Kling M, Entholzner E, Stemberger A, Hipp R: A comparison of the ROTEG coagulation analyser and conventional thromboelastographic systems Anesth Analg 1997; 84:S231.
15 Coakley M, Reddie K, Mackie I, Mallett S: Transfusion triggers in orthotopic liver transplantation: a comparison of ROTEM, thromboelastography and conventional coagulation tests J Cardiothor Vasc Anesth 2006;20:548–553.
15 Lang T, Bauters A, Braun SL, Potzsch B, von Pape KW, Kolde HJ, Lakner M: Multi-centre investigation on reference ranges for ROTEM thromboelastometry. Blood Coagul Fibrinolysis 2005;16:301–310.
17 Görlinger K. Differenzierte Therapie komplexer Gerinnungsstörungen J Anästh Intensivbeh 2005;1: 120–124:
18 Görlinger K. ROTEM – erweitertes perioperatives Gerinnungsmanagement Hämostaseologie 2006;26 (suppl 1):S64–S76.
19 Görlinger K: Gerinnungsmanagement bei Lebertransplantationen. Hämostaseologie 2006;26(suppl 1):S64–S76.
20 Jennings I, Kitchen DP, Woods TA, Kitchen S, Walker ID: Emerging technologies and quality assurance: the United Kingdom National External Quality Assessment Scheme perspective. Semin Thromb Hemost 2007;33:243–249.