

Tissue Factor-Activated Thromboelastograms in Children Undergoing Cardiac Surgery: Baseline Values and Comparisons

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Activation of clotting with tissue factor (TF) allows rapid evaluation of thromboelastograms but alters the values of thromboelastogram variables. We have performed TF-activated thromboelastograms in 250 children <2 yr old undergoing cardiac surgery to establish baseline values. Five groups were distinguished to evaluate the effects of quantitative deficiencies in coagulation factor levels during infancy: <30 days, 1–3 mo, 3–6 mo, 6–12 mo, and 12–24 mo. Activation of clotting (R and K values) was similar among groups. Infants 1–3 mo of age demonstrated increased clot strength compared with the other groups, a finding similar to previous evaluation of native thromboelastograms. The α and maximum amplitude values were numerically

almost identical in each age group, a unique finding in activated thromboelastograms. Fibrinolysis was similar among groups. We believe that knowledge of baseline TF-activated thromboelastogram variables in young children will be useful in interpreting these thromboelastograms in clinical scenarios, in using these thromboelastograms as part of coagulopathy treatment algorithms, and during the application of more specific thromboelastogram modifiers. Additionally, the similarity of α and maximum amplitude values in each age group will allow even faster interpretation of thromboelastogram data.

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The use of thromboelastography to monitor the coagulation system or to guide the treatment of coagulopathies has gained popularity in many areas of medicine in recent years (1,2). Recently described modifications of the thromboelastogram have allowed it to become a more practical coagulation monitor for acute clinical situations. These modifications can be very useful during certain surgical procedures in which timely coagulation data can facilitate the effective management of significant coagulopathies.

In the pediatric cardiac operating room (OR), thromboelastographic data have been shown to correlate with the extent of postoperative blood loss (3–6). However, two obstacles have had to be overcome for thromboelastography to become useful in this setting. First, baseline thromboelastogram variables for infants

and young children needed to be established because the levels of many coagulation factors and inhibitors are significantly lower in young infants compared with adults (7). Thromboelastography in children <2 yr of age has shown functional integrity of their coagulation systems despite these quantitative deficiencies. Baseline thromboelastogram values in young infants differ from those of adults but reference values have been defined for each variable using native blood samples in various age groups of young children (8). Second, blood samples had to be modified to allow thromboelastogram data to be obtained fast enough to be of value in the OR. Activating coagulation by mixing blood with celite or tissue factor (TF) permits the rapid attainment of the α angle and maximum amplitude (MA) variables that correlate with blood loss after cardiopulmonary bypass (CPB) in children (9,10). Adding heparinase or protamine to blood neutralizes circulating heparin and allows thromboelastograms to be run even while patients are anticoagulated during CPB (10–12). The combination of these two modifications has allowed thromboelastography to become a timely coagulation monitor in the cardiac OR.

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To constructively use thromboelastogram modifiers to direct coagulopathy management in the OR, baseline thromboelastogram variables must be reassessed while using the modifiers. This is especially true in young children because of their quantitative coagulation deficiencies. The use of celite or TF to activate the coagulation system results in thromboelastogram values that differ from those obtained from unactivated, native blood (10,13). Because initial clot formation is accelerated, the thromboelastogram variables that reflect the rapidity of clotting, the R and K values, have been found to be significantly shortened compared with those obtained from native blood. Additionally, because clot strength is enhanced after blood activation, the variables that define this strength, the α angle and the MA, are significantly increased. In light of these alterations, the purpose of this descriptive study was to reevaluate thromboelastography using activated blood samples in young infants and children undergoing cardiac surgery and to define baseline values for thromboelastogram variables from activated blood in these young children. We have focused on the use of TF for coagulation activation because clot formation occurs more rapidly after adding TF versus celite to blood samples and because TF activation is being used when the thromboelastogram effects of other coagulation modifiers, such as heparinase and platelet glycoprotein IIb/IIIa receptor inhibitor, are being studied (10,14). Additionally, we have focused on children undergoing cardiac surgery because of the potential clinical usefulness of thromboelastography in managing the coagulopathies that occur after CPB in these children.

Methods

After approval from our IRB, 250 children under 2 yr of age scheduled for elective cardiac surgery were enrolled to obtain 50 children in each of 5 age groups: Group I, younger than 30 days; Group II, 1-3 mo; Group III, 3-6 mo; Group IV, 6-12 mo; Group V, 12-24 mo. None of the enrolled children were receiving any medication known to have an effect on coagulation, none had medical issues other than their congenital heart defect, and none were hemodynamically unstable.

After the induction of general anesthesia and placement of invasive monitoring lines, a 1-mL sample of blood was drawn from the arterial line after aspirating 5 mL of blood to clear the line of heparin from the flush system. A 0.35-mL aliquot of this sample was immediately placed into a preheated disposable cup of a Thrombelastograph[®] coagulation analyzer (Haemoscope Corporation, Skokie, IL) containing 10 μ L of 1% TF. The pin of this thromboelastogram channel was raised and lowered into

the cup five times to ensure adequate mixing of the blood and TF, and a layer of mineral oil was applied to the top of the blood in the cup to prevent evaporation. The thromboelastogram tracing was then begun and allowed to run until at least 60 min after achieving its MA. Subsequently, each tracing was analyzed by manual measurement.

Five values were measured from the thromboelastogram tracing and three indices were calculated using these measured values. The R value (in millimeters), measured from the beginning of the tracing until an amplitude of 2 mm is reached, represents the time necessary for initial clot formation. The K value (in millimeters) is the time interval from the end of the R value until an amplitude of 20 mm is attained and appraises the rapidity of fibrin build-up and cross-linking as the clot forms. The α value similarly assesses this rate of clot formation, is measured as the slope of the outside divergence of the tracing from the point of the end of the R value, and reflects the function of both fibrinogen and platelets. The MA value is a reflection of the maximum strength of the fibrin clot and is influenced most importantly by fibrinogen levels, platelet numbers, and platelet function as well as by factors VIII and XIII. The A-60 value is the amplitude of the tracing 60 min after the MA has been reached and is useful in measuring clot retraction or lysis by comparing it with the MA value. An A-60/MA ratio (whole blood clot lysis index) of <0.85 has been used to define fibrinolysis (15). The shear modulus ($G = [5000]MA/[100 - MA]$) (16) and the coagulation index ($CI = -[0.1227]R + [0.0092]K + [0.1655]MA - [0.0241]\alpha - 5.0020$) (Thrombelastograph[®] Operations Manual and CTEG User's Guide; Haemoscope Corporation) were also calculated because these indices are sensitive measures of actual clot strength and the entire scope of the coagulation process.

Analysis of variance was used to ascertain whether differences existed in the thromboelastograms between groups. Comparisons of groups were then made using two-sided *t*-tests assuming unequal variances with Bonferroni correction for multiple comparisons. χ^2 analysis was used to determine differences between groups in the occurrence of fibrinolysis.

Results

The cardiac lesions present in the children of each age group are delineated in Table 1. The results of our thromboelastogram measurements and indices calculations are compiled in Tables 2 and 3 and are expressed as mean \pm sd. Although R and K values are slightly longer in neonates (Group I), there were no statistical differences among the groups in these values (Table 2). Infants 1-3 mo of age (Group II) demonstrated greater clot

Table 1. Cardiac Diagnoses by Age Group

	<1 mo	1-3 mo	3-6 mo	6-12 mo	12-24 mo
Secundum ASD	—	—	—	1	5
VSD	3	19	12	15	12
AVSD	—	8	12	7	4
TOF	3	8	12	7	9
TGA	8	3	—	2	1
Truncus arteriosus	2	2	—	—	—
APVC	—	1	1	—	1
Tricuspid atresia	4	—	3	2	1
Pulmonary atresia	4	2	1	1	2
Single ventricle ^a	14	2	3	7	6
MS/MR	—	—	—	3	2
AS	—	1	1	1	3
PS	—	—	1	3	1
ALCAPA	—	—	1	1	—
Coronary artery fistula	1	—	—	—	—
AP window	1	—	—	—	—
PDA	—	—	2	—	2
IAA	2	—	1	—	—
Coarctation	7	2	—	—	—
Vascular ring	1	1	—	—	—
Heart block	—	1	—	—	1

ASD = atrial septal defect, VSD = ventricular septal defect, AVSD = atrioventricular septal defect, TOF = tetralogy of Fallot, TGA = transposition of the great arteries, APVC = anomalous pulmonary venous connection, MS = mitral stenosis, MR = mitral regurgitation, AS = aortic stenosis, PS = pulmonary stenosis, ALCAPA = anomalous left coronary artery from the pulmonary artery, AP = aortopulmonary, PDA = patent ductus arteriosus, IAA = interrupted aortic arch. ^a "Single ventricle" used to group children requiring a Norwood procedure initially. Diagnoses included hypoplastic left heart syndrome, unbalanced AVSD, and double inlet left ventricle.

Table 2. Tissue Factor-Activated Thromboelastogram Values

Group	R (mm)	K (mm)	R + K (mm)	α (°)	MA (mm)	A-60 (mm)
I (<30 d)	2.6 ± 4.8	4.5 ± 6.2	7.2 ± 10.8	63.0 ± 13.6	65.9 ± 7.8	60.0 ± 7.9
II (1-3 mo)	1.8 ± 3.5	2.7 ± 1.9	4.5 ± 5.3	72.0 ± 8.0*	72.7 ± 5.4†	66.8 ± 7.3‡
III (3-6 mo)	1.9 ± 4.8	2.7 ± 1.4	4.6 ± 5.5	68.6 ± 9.8	69.0 ± 7.2	63.9 ± 7.3
IV (6-12 mo)	2.7 ± 6.2	3.7 ± 2.3	6.4 ± 8.0	64.3 ± 11.5	67.2 ± 8.1	61.1 ± 8.2
V (12-24 mo)	1.1 ± 1.5	3.1 ± 1.8	4.3 ± 2.9	66.2 ± 9.5	68.3 ± 6.3	62.4 ± 7.5

Values expressed as mean ± SD.
MA = maximum amplitude, A-60 = amplitude 60 min after MA obtained.
* $P < 0.05$ versus Groups I, IV, and V.
† $P < 0.05$ versus Groups I, III, IV, and V.
‡ $P < 0.05$ versus Groups I and IV.

Table 3. Tissue Factor-Activated Thromboelastogram Indices

Group	G (dynes/cm ²)	CI	WBCLI <0.85 (% of patients with fibrinolysis)
I (<30 d)	10,441 ± 3663	4.18 ± 1.35	4
II (1-3 mo)	14,158 ± 4393*	5.18 ± 0.93*	6
III (3-6 mo)	11,950 ± 3710	4.63 ± 1.29	4
IV (6-12 mo)	11,166 ± 4068	4.34 ± 1.41	6
V (12-24 mo)	11,427 ± 3601	4.66 ± 0.88	6

Values expressed as mean ± SD.
G = elastic shear modulus, CI = coagulation index, WBCLI = whole blood clot lysis index (A-60/maximum amplitude).
* $P < 0.05$ versus Groups I, IV, and V.

strength than other children as indicated by significantly larger MA values than those of all 4 other groups as well as by significantly larger α values and shear modulus and coagulation index calculations than those of groups

I, IV, and V (Tables 2 and 3). Interestingly, the numerical values of α and MA in each age group were nearly identical (Table 2). The incidence of thromboelastogram-defined fibrinolysis was similar among groups (Table 3).

Discussion

Our study is the first to report reference values for TF-activated thromboelastogram variables in age-defined subsets of children less than two years of age undergoing cardiac surgery. We chose to study children of this age in order to evaluate the functional effects on activated thromboelastograms of the quantitative deficiencies in coagulation factor levels that occur during infancy (7) and because reference values for native thromboelastograms in the same age-defined subsets of children have been previously described (8). Children undergoing cardiac surgery were selected because they represent a patient population in whom thromboelastography may be of significant clinical use. After TF-activation, R and K values are shorter and α and MA values are larger than values from native blood. In contrast to native thromboelastograms, R and K values after TF activation did not differ among the five age groups. Comparisons of the TF-activated thromboelastograms in the age subsets showed greater clot strength in infants one to three months of age, a finding similar to that seen in thromboelastograms from native blood. A striking similarity was found between the numerical values of α and MA in each age group subset when using TF activation. This finding is different from that seen in native thromboelastogram tracings and could allow even faster interpretation of the coagulation status using TF-activated thromboelastography because decisions need not wait on attainment of the MA value. Thromboelastogram-defined fibrinolysis was seen infrequently (4%–6% of children) but with similar incidence among the age groups after TF activation.

We believe these data are important for several reasons. Baseline “normal” values for TF-activated thromboelastograms must be known to prevent errors in coagulopathy treatment when using these values to direct clinical management. A recent investigation has found that the greatest asset of thromboelastography in cardiac surgery is its “negative predictive value;” i.e., in the face of “normal” thromboelastogram α and MA values after CPB, continuing bleeding is likely not attributable to a persistent coagulopathy but to a surgical cause (17). Therefore, it must be recognized that baseline α and MA values are larger after TF activation of blood in order to prevent the misinterpretation of abnormally small activated values as an indication of a corrected coagulopathy and the need for surgical re-exploration.

The use of thromboelastogram-guided transfusion algorithms in the OR after cardiac surgery in adults has been shown to reduce blood product administration and blood loss compared with situations in which routine clotting tests have been used to decide upon interventions (18). The use of similar algorithms has been considered in children. Activation of coagulation

by TF will allow the most rapid attainment of thromboelastogram data for these algorithms (10). However, clinicians again must be aware of normal TF-activated thromboelastogram values in order to define appropriate trigger levels for blood product administration. We have defined these reference values for children less than two years old undergoing cardiac surgery. Similar data for celite-activated thromboelastograms in children undergoing noncardiac surgery have been published previously although the age groupings were different than those we have used (9).

The definition of baseline TF-activated thromboelastogram values in young children provides a foundation on which further, more specific thromboelastogram modifications can be based. The use of heparinase or protamine, for instance, allows the acquisition of coagulation data during the anticoagulated state of CPB (10–12). The addition of a platelet glycoprotein IIb/IIIa receptor inhibitor has been found to delineate the contribution of platelets and fibrinogen to clot strength in adults (14). Because the pertinent thromboelastogram variables sought after the addition of these modifiers are the α and MA values, TF activation is useful in expediting these thromboelastograms yet the baseline values for α and MA after TF activation must be appreciated.

Two findings in the TF-activated thromboelastogram data differ from what has previously been noted in native thromboelastograms of the same-aged children. First, there are no significant differences in the R and K values among the groups after TF activation (Table 2) whereas, in native thromboelastograms, the K values of infants one to three months (Group II) and three to six months (Group III) were significantly shorter than those of the other age groups (8). Initial models of coagulation based on intrinsic and extrinsic cascades are being supplemented by a cell-based model of coagulation that emphasizes the substantial role of TF in initiating thrombin generation and subsequent fibrinogen cleavage (19). The recently appreciated pivotal role of TF in stimulating coagulation may explain the similarity in the onset of clotting across all of the age groups in our current study. This finding also serves to reemphasize the functional integrity of the activated coagulation systems of neonates and young infants despite their quantitative deficiencies in the coagulation factors that have roles in the initial steps of blood clotting.

Second, the α and MA values from TF-activated thromboelastograms are numerically similar in each age group. Whereas the α value is indicative of the rate of clot formation and the MA is reflective of clot strength, both are significantly influenced by both fibrinogen and platelets (15). The intensity of the stimulation of coagulation by TF and the resulting burst of

thrombin generation would rapidly activate both fibrinogen and platelets and would amplify the resulting α and MA thromboelastogram values (19). The resulting numerical equality of these two variables should permit even faster interpretation of a child's coagulation status because the α value is known within 2-4 minutes of initiation of a TF-activated thromboelastogram whereas 10-15 more minutes are required for the MA to become evident on the thromboelastogram tracing (data not shown).

A shortcoming of this study is the lack of concurrently run native and TF-activated thromboelastograms for direct comparison. However, a previous study by our group defined native thromboelastogram values in a large group of similarly age-stratified children less than two years old (8). Because the children in both studies were in overall good health and were presenting for elective surgery and because the thromboelastogram data for both of these studies were acquired using the same Thrombelastograph[®] coagulation analyzer machines, we believe the data from the two studies can be compared. Indeed, the finding of thromboelastogram values indicative of a more coagulable state in infants one to three months of age (Group II) and the finding of no significant differences in any variables among Groups I, IV, and V are identical in both studies. Additionally, the study population in the current investigation was children with congenital heart disease (CHD) undergoing cardiac surgery. Because some of these children may possess baseline coagulation alterations not present in children without CHD, care must be taken in generalizing our data to other patient groups. However, the similarities mentioned above between our current findings and those of native thromboelastograms in similarly aged children without CHD may allay this concern.

In summary, we have evaluated the effect of TF activation on thromboelastograms in age-defined subsets of children less than two years old undergoing cardiac surgery and have defined reference values for TF-activated thromboelastogram variables in these subsets of children. The use of these data will allow accurate interpretation of TF-activated thromboelastograms in clinical situations of altered coagulation and will provide a foundation on which further modifications of thromboelastography can be investigated to help define and treat acute coagulopathies in similarly aged children.

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