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# Effect of Fibrinogen Concentrate on Intraoperative Blood Loss Among Patients With Intraoperative Bleeding During High-Risk Cardiac Surgery

## A Randomized Clinical Trial

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 Supplemental content

**IMPORTANCE** Fibrinogen concentrate might partly restore coagulation defects and reduce intraoperative bleeding.

**OBJECTIVE** To determine whether fibrinogen concentrate infusion dosed to achieve a plasma fibrinogen level of 2.5 g/L in high-risk cardiac surgery patients with intraoperative bleeding reduces intraoperative blood loss.

**DESIGN, SETTING, AND PARTICIPANTS** A randomized, placebo-controlled, double-blind clinical trial conducted in Isala Zwolle, the Netherlands (February 2011-January 2015), involving patients undergoing elective, high-risk cardiac surgery (ie, combined coronary artery bypass graft [CABG] surgery and valve repair or replacement surgery, the replacement of multiple valves, aortic root reconstruction, or reconstruction of the ascending aorta or aortic arch) with intraoperative bleeding (blood volume between 60 and 250 mL suctioned from the thoracic cavity in a period of 5 minutes) were randomized to receive either fibrinogen concentrate or placebo.

**INTERVENTIONS** Intravenous, single-dose administration of fibrinogen concentrate (n = 60) or placebo (n = 60), targeted to achieve a postinfusion plasma fibrinogen level of 2.5 g/L.

**MAIN OUTCOMES AND MEASURES** The primary outcome was blood loss in milliliters between intervention (ie, after removal of cardiopulmonary bypass) and closure of chest. Safety variables (within 30 days) included: in-hospital mortality, myocardial infarction, cerebrovascular accident or transient ischemic attack, renal insufficiency or failure, venous thromboembolism, pulmonary embolism, and operative complications.

**RESULTS** Among 120 patients (mean age; 71 [SD, 10] years, 37 women [31%]) included in the study, combined CABG and valve repair or replacement surgery comprised 72% of procedures and had a mean (SD) cardiopulmonary bypass time of 200 minutes (83) minutes. For the primary outcome, median blood loss in the fibrinogen group was 50 mL (interquartile range [IQR], 29-100 mL) compared with 70 mL (IQR, 33-145 mL) in the control group ( $P = .19$ ), the absolute difference 20 mL (95% CI, -13 to 35 mL). There were 6 cases of stroke or transient ischemic attack (4 in the fibrinogen group); 4 myocardial infarctions (3 in the fibrinogen group); 2 deaths (both in the fibrinogen group); 5 cases with renal insufficiency or failure (3 in the fibrinogen group); and 9 cases with reoperative thoracotomy (4 in the fibrinogen group).

**CONCLUSIONS AND RELEVANCE** Among patients with intraoperative bleeding during high-risk cardiac surgery, administration of fibrinogen concentrate, compared with placebo, resulted in no significant difference in the amount of intraoperative blood loss.

**TRIAL REGISTRATION** clinicaltrials.gov Identifier: [NCT01124981](https://clinicaltrials.gov/ct2/show/study/NCT01124981) and EudraCT No: [2009-018086-12](https://eudract.eu/number/2009-018086-12)

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Excessive bleeding is one of the most common complications in cardiac surgery with approximately 15% to 20% of patients consuming more than 80% of all blood products used in cardiac surgery procedures.<sup>1</sup> Bleeding may result in a need for red blood cell transfusion. Because these transfusions increase the risk of adverse outcomes, avoiding them is preferable.<sup>2,3</sup> Intraoperative bleeding during cardiac surgery is frequently treated with coagulation factor replacement therapies, but their efficacy has not been definitively established.

Bleeding during cardiac surgery might be influenced by patient- and procedure-related factors, use of anticoagulant medication, inflammation, consumption and dilution of clotting factors, and fibrinolysis associated with the use of cardiopulmonary bypass (CPB).<sup>4,5</sup> When major blood loss is substituted with red blood cells and colloid fluids, plasma fibrinogen deficiency develops earlier than any other clotting factor,<sup>6</sup> which may lead to excessive postoperative blood loss requiring blood products transfusions.<sup>7,8</sup> Plasma fibrinogen (factor I) plays a key role in hemostasis by acting as an endogenous substrate for fibrin formation, promoting clot formation and platelet aggregation by binding platelet glycoprotein IIb/IIIa receptors.<sup>9,10</sup> Because of fibrinogen's key role in maintaining hemostasis and because it is rapidly depleted during cardiac surgery, fibrinogen replacement therapy has gained popularity in treating cardiac surgery-associated bleeding.<sup>11</sup>

Infusion of fibrinogen concentrate replaces depleted plasma fibrinogen and reverses coagulopathic bleeding by enhancing the speed and strength of blood clot formation, which should decrease intraoperative bleeding.<sup>12,13</sup> Fibrinogen concentrate infusion is given to control bleeding during cardiac surgery but, to our knowledge, only 2 randomized clinical trials have investigated its use in cardiac surgery. One of these studies enrolled a total of 60 patients, and the other examined fibrinogen concentrate infusion given in doses based on thromboelastometry measurement but not related to ongoing blood loss.<sup>14,15</sup> Neither of these studies provided definitive evidence for the efficacy of fibrinogen concentrate infusion to control ongoing bleeding during cardiac surgery.

The current study sought to determine whether fibrinogen concentrate infusion reduced ongoing blood loss in patients undergoing cardiac surgery if it was administered with the intent of achieving a post-infusion plasma fibrinogen level of at least 2.5 g/L.

## Methods

### Study Design and Study Population

This study was a randomized, placebo-controlled, double-blind clinical trial of fibrinogen concentrate (Hemocomplettan P, CSL Behring) vs placebo for the treatment of intraoperative bleeding during high-risk cardiac surgery. The study was conducted in the Isala Zwolle, a large university-affiliated teaching hospital in the Netherlands. The study protocol was approved on August 23, 2010, by the institutional medical ethical committee and was conducted in accordance with the prin-

### Key Points

**Question** What is the effect of fibrinogen concentrate on blood loss in patients with intraoperative bleeding during high-risk cardiac surgery?

**Findings** In this randomized clinical trial involving 120 patients undergoing cardiac surgery with intraoperative bleeding, administration of fibrinogen concentrate, compared with placebo, resulted in no significant difference in the amount of intraoperative blood loss.

**Meaning** Among patients with intraoperative bleeding during high-risk cardiac surgery, fibrinogen concentrate infusion did not reduce intraoperative blood loss.

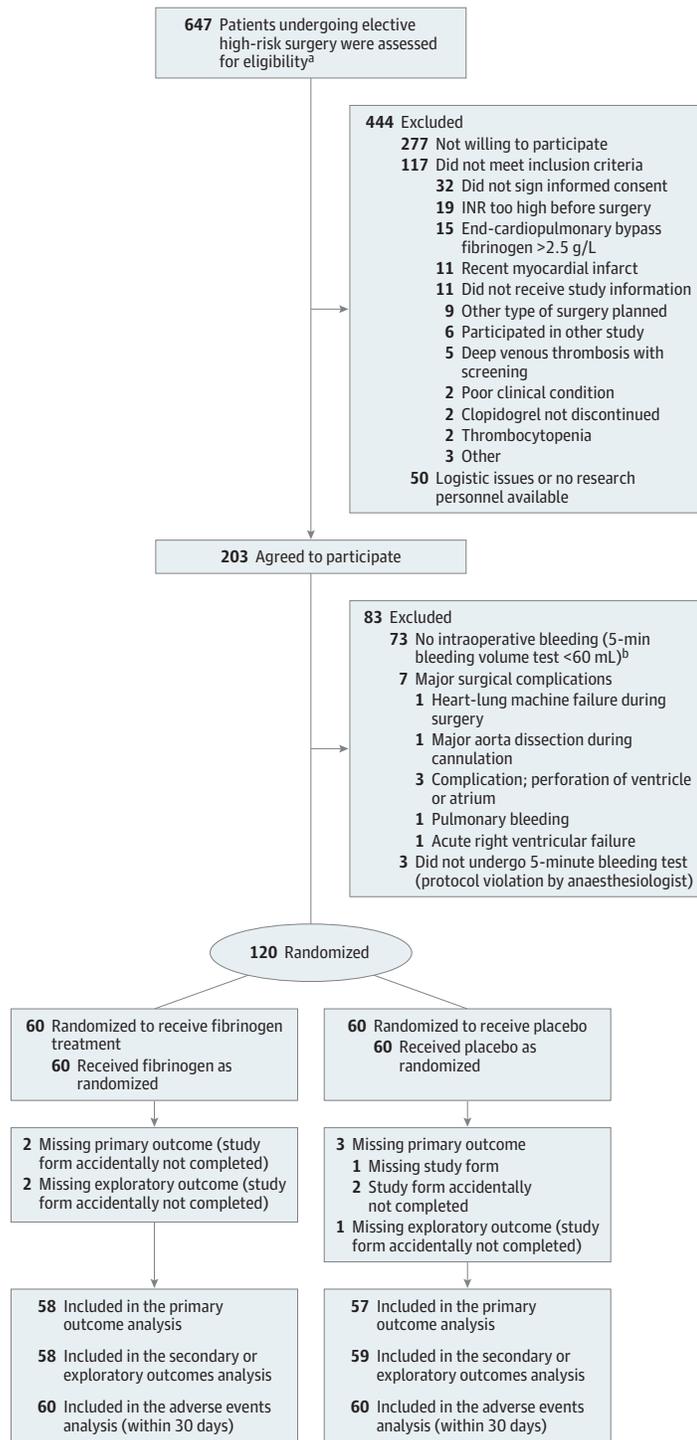
ciples of the Declaration of Helsinki and the International Conference on Harmonisation of Good Clinical Practice guidelines. Recruitment of patients occurred between February 2011 and December 2014. On February 2011 the protocol was amended with minor changes. The full study protocol and the amendment are available in [Supplement 1](#).

Patients were eligible if they were older than 18 years and had undergone elective high-risk cardiac surgery defined as combined coronary artery bypass graft (CABG) surgery and valve repair or replacement surgery, the replacement of multiple valves, aortic root reconstruction, or reconstruction of the ascending aorta or aortic arch (**Figure 1**). Specific exclusion criteria were proof or suspicion of a congenital or acquired coagulation disorder, any stroke or myocardial infarction within 2 months preceding surgery, venous or arterial thrombosis, clopidogrel use in the 5 days preceding surgery, glycoprotein IIb/IIIa receptor antagonist use in the 2 days preceding surgery, or INR (international normalized ratio) of more than 1.4 in patients who used coumarins. Eligible patients were informed and asked for a signed informed consent a week prior to the surgery date with a patient information letter that had been approved by the institutions' medical ethical committee. One day prior to surgery, trained research nurses screened the eligible patients for participation in the study.

The study data were prospectively collected starting a day prior to surgery and ending at the 30-day follow-up with collection of information on adverse events by sending a questionnaire regarding their clinical condition and, if necessary, by contacting the referring physician. The referring physician was contacted in case the patient did not return the follow-up form after it was sent for the third time. When the patient had documented an event in the form, the referring physician was also contacted for more details of the event.

During the entire study period, anesthetic management and surgical treatment was performed according to the same standard procedures as described previously.<sup>16</sup> Antifibrinolytic prophylaxis was given to all study participants (ie, both the fibrinogen and control groups) as part of the surgical procedure. After the infusion of study medication, the criteria for transfusion were based on a transfusion protocol introduced at the Isala Zwolle in the year 2009, specifically designed for cardiac surgery procedures.<sup>17</sup> In short,

Figure 1. Flow Diagram of Progress Through Phases of a Randomized Trial Comparing Fibrinogen Concentrate vs Placebo for Intraoperative Bleeding Among Patients Undergoing High-Risk Cardiac Surgery



MRSA indicates methicillin-resistant *Staphylococcus aureus*.

<sup>a</sup> High-risk cardiac surgery indicates combined coronary artery bypass graft surgery and valve repair or replacement surgery, the replacement of multiple valves, aortic root reconstruction, or reconstruction of the ascending aorta or aortic arch.

<sup>b</sup> No intraoperative bleeding occurred when "5-minute bleeding volume test" by suctioning of blood from the thoracic cavity (pleura and pericardium) for a period of 5 minutes revealed collected volumes of less than 60 mL; these patients were excluded.

1 unit of red blood cells was administered when the hemoglobin (Hb) level was lower than 8.5 g/dL. Patients received 2 units of fresh frozen plasma when they lost at least 1 liter of plasma, and when they lost 2 or more liters of plasma, patients received 4 units of fresh frozen plasma via transfusion. A platelet count of less than  $80 \times 10^9/L$  was transfused

with 1 unit of platelet concentrate (5 donors). Pursuant to the intensive care unit (ICU) protocol, patients experiencing a blood loss exceeding 400 mL in 1 hour or exceeding 300 mL per hour for 2 hours or exceeding 200 mL per hour for 3 hours while in the ICU received 4 units of red blood cells, 4 units fresh frozen plasma, and 1 unit of platelet con-

concentrate (5 donors) via transfusion and the attending surgeon was informed. Patients whose platelet counts were lower than  $75 \times 10^9/L$  received 1 unit of platelet concentrate (5 donors) via transfusion.

### Intervention

Reconstitution of fibrinogen concentrate and formulation of the placebo were carried out under aseptic conditions by trained research personnel. Solutions that were cloudy or contained residues (deposits or particles) were not used. Fibrinogen concentrate was diluted in 50 mL of sterile water (room temperature). For administration of placebo, 2 g of albumin (albuman, 200 g/L, Sanquin CLB) was diluted with 0.9% of sodium chloride (room temperature) and placed into a 50 mL syringe. All study medication was administered through an 18-gauge peripheral intravenous line in the brachial vein.

When cardiopulmonary bypass was completed, hemostasis of the operative field was attempted by using electrocautery, packing, and suture repair of any bleeding occurring at anastomotic sites. Then intraoperative bleeding was measured by a 5-minute bleeding volume test if the following conditions were met: activated clotting time of less than 140 seconds, body temperature higher than  $36^\circ C$ , blood pH higher than 7.30, and an Hb concentration higher than 8.5 g/dL ( $>5.3$  mmol/L), or hematocrit higher than 25%. Bleeding was measured by packing the thoracic cavity with fresh surgical gauze and allowing exactly 5 minutes to pass before they were removed. The gauze was then wrung out and weighed. Blood loss that was collected from suctioning the operative field was measured using a separate collector with a scale accurate to the milliliter connected to the cell saver. Blood volumes in the collector were read by 2 operating room personnel, and the reported volume was the value that these personnel agreed on. The total blood loss was measured by adding the amount of blood that was in the sponges to that measured from suctioning of the thoracic cavity.

Bleeding volumes less than 60 mL were considered "dry" and classified as having no intraoperative bleeding; these patients were excluded from the study. Bleeding volumes more than 250 mL were classified as having surgical bleeding. These patients had additional efforts at achieving surgical hemostasis such as electrocautery, packing, and suture repair of any bleeding vascular anastomoses. This was followed by repetition of the 5-minute bleeding volume test. Bleeding volumes between 60 mL and 250 mL were classified as having intraoperative bleeding. The method we used for measuring intraoperative bleeding was also used in previous studies.<sup>18,19</sup> When the 5-minute bleeding volume test indicated intraoperative bleeding, patients were randomized to receive fibrinogen concentrate or placebo. Fibrinogen doses were calculated,<sup>12,18-22</sup> based on plasma fibrinogen levels at the end of cardiopulmonary bypass surgery measured with the Clauss method (turbidometric assay).<sup>23</sup> The postinfusion target plasma fibrinogen concentration was 2.5 g/L. The intervention was considered to have started at the initiation of the study medication infusion (fibrinogen concentrate or placebo), which was given

after cardiopulmonary bypass was completed and intraoperative bleeding was established. The formula used for dosing of fibrinogen concentrate was

$$(2.5 - [\text{Plasma Fibrinogen Level at the End of Cardiopulmonary Bypass, g/L}]) \times 0.07 \times (1 - \text{Hematocrit on Cardiopulmonary Bypass}) \times \text{Body Weight (kg)} = \text{Whole Grams Fibrinogen Concentrate To Be Dosed}$$

This formula was used in earlier studies with point-of-care thromboelastometry variables (fibrinogen trace) to determine the dose of fibrinogen concentrate for target plasma fibrinogen levels.<sup>13,18,19</sup> In this trial, the plasma fibrinogen concentrations were used to calculate the dose of fibrinogen concentrate. The circulating plasma volume was estimated from body weight and corrected for hematocrit.

Patients with a plasma fibrinogen level of more than 2.5 g/L at the end of cardiopulmonary bypass (which results in the formula yielding a negative value) were excluded from the study because they already had fibrinogen levels greater than what was targeted for this study.

Fibrinogen concentrate was administered as a single-dose intravenous infusion. Postinfusion plasma fibrinogen concentrations were measured immediately following ICU admission. All study and clinical personnel were blinded to the measured postinfusion plasma fibrinogen concentrations (the results were not shown in clinical laboratory results available to the treatment teams) until official unblinding of the trial. All observers were blinded to the study drug assignment.

### Study Outcomes

The primary outcome of this study was intraoperative blood loss (mL) measured between intervention (ie, infusion of study medication after completion of cardiopulmonary bypass) and closure of the chest when the surgery ended.

Secondary outcomes included the measured blood loss 1, 3, 6, 12, and 24 hours after the intervention. Other secondary outcomes were the proportion of patients who had received a transfusion and the number of units of red blood cells, fresh-frozen plasma, platelet concentrate, and any blood product units used between when the intervention started and when the chest was closed and for the 24-hour period after the intervention. The amount of procoagulants and antifibrinolytics (tranexamic acid, desmopressin, prothrombin complex concentrate, recombinant factor VIIa and fibrinogen concentrate) given during the perioperative and postoperative periods was also assessed.

To evaluate safety and tolerability of fibrinogen concentrate, adverse events were assessed as a secondary analysis. These included 30-day in-hospital mortality, myocardial infarction, cerebrovascular accident or transient ischemic attack, renal insufficiency or failure, venous thromboembolism, pulmonary embolism, allergic or other systemic reaction to study medication, and sternal or wound infections and reoperative thoracotomy (within 5 days of initial surgery). Thrombosis in the lower extremity was diagnosed with compression ultrasound testing 1 day before surgery and at day 3 days after surgery.<sup>24,25</sup>

### Randomization and Blinding

The participants were randomized with a web-based randomization protocol using an unstratified fixed-block size of 4. The order of blocks was also randomized. Much attention was given to ensure strict blinding during the randomization process, the infusion of study medication (placebo or fibrinogen concentrate), the follow-up period, and during data collection. The medical team involved in the entire surgical process (ie, in the operating room, ICU, and ward) were blinded for the allocated treatment. The following measures were taken to ensure blinding of the treatment allocation; the room where the allocation was performed had restricted access to only research team members who could enter it only by using a personal badge. Only research team members had access to the web-based randomization protocol and were authorized to prepare the study medications. To avoid revealing the treatment allocation by study drug volume, temperature, color, or viscosity, the infusates were prepared for both the treatment and placebo groups using the same dosing formula and were delivered in amber colored syringes (50 mL, Luer-Lok BD Plastipak). Diluted albumin was used as a placebo and had the same amount of protein (2 g in 50 mL) as did the fibrinogen preparation. During the first 24 hours after infusion of study medication, measured plasma fibrinogen concentration results were not revealed until official unblinding of the trial, which occurred at the final stage of the statistical analysis.

### Sample Size Calculation

The sample size was calculated based on prior observations of the total intraoperative blood loss during cardiac surgery (ie, from start of surgery to closure of the chest).<sup>26</sup> The median total intraoperative blood loss (including blood loss as measured with the cell saver) was 2200 mL. It was expected that the anticipated total intraoperative blood loss of 2200 mL could be reduced by 40% to 1350 mL. The anticipated 40% reduction in blood loss attributable to fibrinogen infusion was based on earlier porcine and human cardiac surgery studies.<sup>18,19,27</sup> Using a power of 80% and an overall level of significance of 0.05, 53 patients were required in each group, resulting in 106 patients in total. To account for potential loss to follow-up, 120 participants were included in the study.

### Statistical Analysis

The mean and standard deviations were calculated for normally distributed continuous variables and the median with interquartile range (IQR) were calculated for nonnormally distributed variables. The statistical analysis plan is available in [Supplement 2](#). Normality was determined with the Kolmogorov-Smirnov statistical test. Frequencies with percentages were calculated for categorical variables. Nonnormally distributed primary and secondary blood loss outcomes were  $\log_{10}$ -transformed and both were presented using medians with IQRs. Missing data for the primary outcome were assumed missing completely at random and were excluded from the analysis. The analysis was repeated using the multiple-imputation approach described in the [Appendix in Supplement 3](#).

For estimating differences between the fibrinogen group and the control group, bivariable linear regression analysis was used with log-transformed blood loss as the dependent variable and treatment as the only (dichotomous) covariate. Given the small size of the study and possible remaining differences between groups after randomization, an exploratory multivariable linear regression analysis was performed on the primary outcome adjusting for age, sex, EuroSCORE (numerical),<sup>28</sup> lowest core temperature, and cardiopulmonary bypass time.

Between-group comparisons of the prespecified secondary outcome, blood loss at the ICU, were made using an exploratory mixed model for repeated measurements analysis. The outcome was the amount of blood loss in each interval, which was log-transformed because of the skewed data. The models included group (ie, fibrinogen or placebo) and time as fixed categorical covariates, and group  $\times$  time interactions. An unstructured covariance matrix was used to account for the correlation of blood loss between time points within individuals because it provided the best model fit as assessed by Akaike information criteria. The results of this exploratory secondary outcome were based on mixed-model between-group comparisons at 1, 3, 6, 12, and 24 hours after chest closure measured in the ICU, using contrasts. No adjustment for multiple comparisons was performed. Missing data for the post hoc exploratory analysis (ie, blood loss at the ICU) were accounted for by a mixed-model regression of repeated measurements. Differences in the proportion of patients who had received a transfusion between the fibrinogen and control groups were calculated, as were the number of units used (red blood cells, fresh-frozen plasma, platelet concentrate, and any blood product) between the fibrinogen and control groups in the first 24 hours following the intervention. In addition, differences in use of procoagulants and antifibrinolytics and clinical adverse event occurrence between the 2 groups were calculated. Statistical significance was assumed if a 2-sided  $P < .05$ .

After 50% of the outcomes were collected, a planned interim analysis of efficacy on blinded data was performed by the data and safety monitoring board. The O'Brien-Fleming stopping criterion was used to control for type I error. The interim analysis used a 2-sided  $\alpha$ -level criteria of .003 (0.3%) and the final analysis applied an  $\alpha$ -level criteria of .049 (4.9%).

All analyses were performed using R 2.15.0 (R Foundation for Statistical Computing; <http://www.R-project.org>) using the base system plus "MICE," "lme4," and "rms" libraries as well as SPSS 21.0 (IBM SPSS Statistic). The statistical analysis plan is available in [Supplement 2](#).

## Results

During the study period from February 2011 to January 2015, 647 patients undergoing elective high-risk cardiac surgery were eligible to participate in the trial (Figure 1). After initial screening, 203 patients agreed to participate and provided informed consent. During surgery, 73 patients had no intraoperative bleeding, 7 had major surgical complications,

Table 1. Baseline and Patient Characteristics

	Fibrinogen (n = 60)	Control (n = 60)
Age, mean (SD), y	70 (10)	72 (10)
Women, No. (%)	21 (35)	16 (27)
Body surface area, mean (SD), m <sup>2</sup>	1.96 (0.19)	1.99 (0.22)
EuroSCORE, mean (SD) <sup>a</sup>	7 (3)	7 (2)
Diabetes type 1 or 2, No. (%)	16 (27)	19 (32)
Hypertension, No. (%)	38 (63)	41 (68)
Left ventricular hypertrophy, No. (%)	27 (45)	32 (53)
Preoperative use of anticoagulants, No. (%)		
Aspirin	31 (52)	33 (55)
Clopidogrel	0 (0)	1 (2)
Nadroparin	8 (13)	6 (10)
Coumarins	16 (27)	10 (17)
Left ventricular ejection fraction %, No. (%)		
<30	9 (15)	5 (8)
30-50	18 (30)	10 (17)
>50	33 (55)	45 (75)
Previous cardiac surgery, No. (%)	5 (8)	4 (7)
Type of surgery, No. (%)		
CABG surgery and valve(s)	43 (72)	43 (72)
Valves only	6 (10)	7 (11)
Thoracic aorta surgery	11 (18)	10 (17)
Preoperative data, mean (SD)		
Hemoglobin, g/dL	14.3 (1.7)	13.9 (1.4)
Hematocrit, %	43 (5)	41 (4)
Platelet count, ×10 <sup>9</sup> /L	222 (78)	220 (53)
End-CPB data, before study medication, mean (SD)		
Hemoglobin, g/dL	9.2 (1.3)	9.2 (1.3)
Hematocrit, %	27 (4)	27 (4)
Platelet count, ×10 <sup>9</sup> /L	131 (37)	142 (39)
Plasma fibrinogen concentration, g/L	1.7 (0.4)	1.8 (0.3)
5-min bleeding volume, median (IQR), mL	75 (66-100)	80 (70-100)
Transfusion during CPB, mean (SD)		
Red blood cells units	0.95 (2.02)	0.61 (1.00)
Fresh-frozen plasma units	0.03 (0.17)	0.00 (0.00)
Transfusion of autologous blood, median (IQR), mL	500 (313-650)	450 (350-638)
Lowest core temperature, median (IQR), °C	30 (28-32)	30 (28-32)
CPB time in minutes, mean (SD)	209 (94)	192 (71)

Abbreviations: CABG, coronary artery bypass graft; CPB, cardiopulmonary bypass; IQR, interquartile range.

<sup>a</sup> EuroSCORE (European System for Cardiac Operative Risk Evaluation) is a risk model for calculating the risk of death after a heart operation using 17 items relating to the patient, state of the heart and the proposed operation. EuroSCORE categorizes risk of death in low risk (EuroSCORE 0-2), intermediate risk (EuroSCORE 3-5), and high risk (EuroSCORE ≥6).

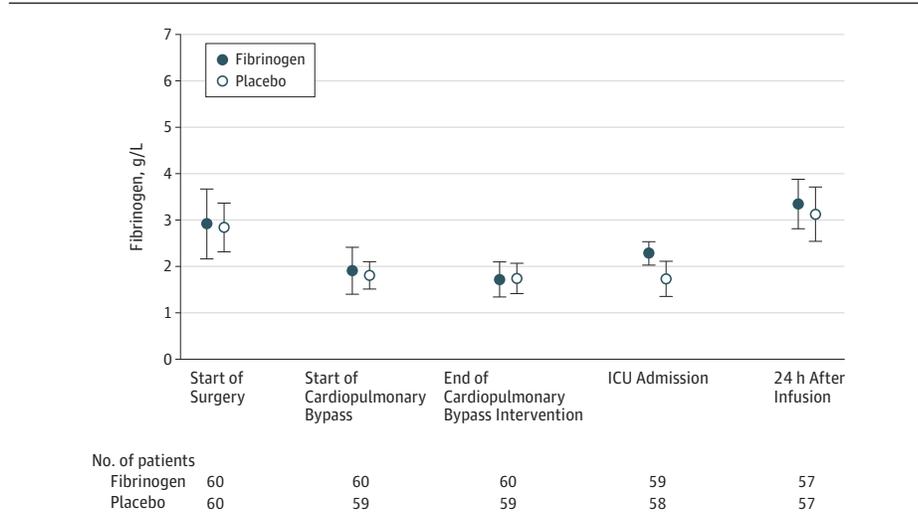
and 3 did not undergo the 5-minute bleeding volume test and were excluded from the study. One hundred twenty patients diagnosed with intraoperative bleeding after removal of cardiopulmonary bypass were randomized to receive placebo or fibrinogen concentrate. In August 2013, an interim analysis was performed on all outcomes (primary and exploratory and safety outcomes). Based on no statistically significant differences between groups, the data and safety monitoring board concluded that the trial should continue.

Among the 120 patients (mean [SD] age; 71 (10) years; 37 women [31%]) included in the study, 72% underwent CABG surgery combined with valve repair or replacement surgery. The mean (SD) cardiopulmonary bypass time was 200 (83) minutes. Table 1 shows the baseline characteristics for each group. The mean preoperative plasma fibrinogen concentration was

2.9 g/L (95% CI, 2.7-3.1 g/L) for the fibrinogen group and 2.8 g/L (95% CI, 2.7-3.0 g/L) for the control group. The mean infused dose of fibrinogen concentrate was 3.1 g (95% CI, 2.7-3.5 g/L) for the fibrinogen group, yielding a mean plasma fibrinogen concentration of 2.3 g/L (95% CI, 2.2-2.4 g/L) as measured when the patients arrived in the ICU. In the control group, the mean plasma fibrinogen concentration at arrival in the ICU was 1.7 g/L (95% CI, 1.6-1.8 g/L). The plasma fibrinogen concentration 24 hours after intervention was 3.3 g/L (95% CI, 3.2-3.5 g/L) for the fibrinogen group and 3.1 g/L (95% CI, 2.9-3.3 g/L) for the control group.

Figure 2 depicts the plasma fibrinogen levels for the first 24 hours following surgery. There were 5 patients with missing data for the primary outcome; 2 in the fibrinogen group and 3 in the control group. These participants were deleted

Figure 2. Mean Plasma Fibrinogen Levels During the Study



Data markers indicate the mean and error bars, standard deviations.

Table 2. Primary, Secondary, and Exploratory Study Outcomes

	Median (IQR), mL		Absolute Difference (95% CI)	P Value
	Fibrinogen (n = 58)	Control (n = 57)		
<b>Primary Outcome</b>				
Blood loss between intervention and chest closure	50 (29-100)	70 (33-145)	20 (-13 to 35) <sup>a</sup>	.19
<b>Secondary or Exploratory Outcome</b>				
No. of patients	58	59		
Blood loss in the ICU/time interval starting from admission				
0-1 h	70 (35-130)	90 (46-149)		
>1-3 h	80 (50-156)	110 (40-220)		
>3-6 h	100 (54-169)	110 (60-208)		
>6-12 h	110 (80-160)	125 (83-224)		
>12-24 h	130 (80-180)	160 (90-270)		
Cumulative 24-h blood loss	570 (390-730)	690 (400-1090)	120 (-45 to 355) <sup>a</sup>	.047 <sup>b</sup>

Abbreviations: ICU, intensive care unit; IQR, interquartile range.

Time point "intervention" is defined as moment of infusion of study medication.

<sup>a</sup> 95% confidence interval of difference in medians is based on a nonparametric bootstrap procedure (10 000 bootstraps with replacement).

<sup>b</sup> P value is based on the constructed mixed-model for repeated measurements.

from the analysis of the primary end point. The 30-day follow-up data regarding clinical adverse events were complete for all 120 patients

**Primary Outcome**

There was no significant difference in blood loss measured from the time of the fibrinogen infusion and chest closure between the fibrinogen group (median, 50 mL; IQR, 29-100 mL) and the control group (median, 70 mL; IQR, 33-145 mL; *P* = .19) with an absolute difference of 20 mL (95% CI, -13 to 35 mL; Table 2). The duration of primary outcome collection was 4.2 minutes (95% CI, 0.4-8.0 minutes) in the fibrinogen group and 8.7 minutes (95% CI, 5.2-12.1 minutes) minutes in the control group. The blood loss difference between the fibrinogen concentrate and control groups was not significantly different after adjustment for the predefined variables age, sex, EuroSCORE, lowest core temperature, and cardiopulmonary bypass time (*P* = .09). Results of a sensitivity analysis based on multiple imputation of missing data were consistent with those of the primary analysis.

**Secondary and Exploratory Analysis**

Transfusion outcomes are shown in Table 3. There were numerically fewer patients and number of units of blood products used in the fibrinogen group compared with the control group. This study was not adequately powered to formally test these secondary outcomes. The same holds for procoagulant and antifibrinolytic use and for adverse events.

Following surgery, 1 patient (2%) in the fibrinogen group and 6 patients (10%) in the control group were given additional fibrinogen concentrate by the treating clinicians. Tranexamic acid was given to 9 patients (15%) in the fibrinogen group and 15 (25%) in the control group. Prothrombin complex concentrate was given to 4 patients (7%) in the fibrinogen group and 10 (17%) in the control group (see eTable 2 in the Supplement 3).

Clinical adverse events occurring during the trial are shown in Table 4. There were more adverse events in the fibrinogen group: 4 strokes or transient ischemic attacks in fibrinogen patients and 2 in the control patients, 3 myocardial infarctions in the fibrinogen patients compared with 1 in

the control patients, and 2 mortalities in the fibrinogen group but none in the control patients. Compression ultrasound of both legs on the third postoperative day showed that no patients had deep venous thrombosis in either group. Compression ultrasound was not performed in 4 patients on day 3. Information on the clinical condition of these patients was obtained within the 30-day follow-up.

Adverse events that occurred during the study along with infused doses of fibrinogen concentrate, fibrinogen plasma concentrations before and after intervention and the time of occurrence of the clinical adverse event in days after surgery are shown in eTable 3 in the and the exact definitions of the clinical adverse events are shown in eTable 1 in Supplement 3

Exploratory analysis using mixed-model regression for repeated measurements showed that cumulative blood loss in the first 24 hours after surgery was significantly lower for the fibrinogen group than for the control group with median blood loss of 570 mL (IQR, 390-730 mL) in fibrinogen patients compared with 690 mL (IQR, 400-1090 mL;  $P = .047$ ) for control patients.

## Discussion

Targeted fibrinogen concentrate infusion in high-risk cardiac surgery did not significantly reduce intraoperative blood loss. More adverse events within 30 days were observed in the fibrinogen group (including stroke and transient ischemic attack, MI, and deaths). Post hoc exploratory analyses suggested a small reduction in cumulative median blood loss (120 mL) in the first 24 hours after surgery, although this difference was not clinically important.

To our knowledge, this study is one of the first to investigate the effects of fibrinogen concentrate on intraoperative blood loss in cardiac surgery procedures. Prior studies either examined the effect of fibrinogen concentrate on other operations or used different clinical end points. For example, in one prior study of the effect of fibrinogen infusion, the procedure was aortic surgery and the outcome was the cumulative amount of all blood products transfused over a 24-hour period.<sup>14</sup> The total amount of infused fibrinogen concentrate in that study was a median of 8 g resulting in a mean (SD) post-intervention plasma fibrinogen concentration of 2.6 (0.5) g/L.

Even though there was less utilization of blood products in the fibrinogen group, at 45 days of follow-up, 1 patient in the fibrinogen group had a myocardial infarction and 2 patients in the control group had a cerebral hemorrhage or infarction. One patient in the fibrinogen group died compared with 4 patients in the control group, resulting in an unclear overall benefit attributable to fibrinogen infusion.

In the current study, plasma fibrinogen concentrations steadily increased following surgery in both the fibrinogen and control groups. Plasma fibrinogen levels were higher in the fibrinogen than in the control group for the first 24 hours following surgery (Figure 2). At 24 hours, plasma fibrinogen levels were nearly equal in the 2 groups: 3.3 g/L for the fibrinogen group and 3.1 g/L for the control group. These results are similar to the aortic surgery randomized clinical trial in which the plasma fibrinogen concentration 1 day after surgery was 3.4 g/L for the fibrinogen group and 3.3 g/L for the control group.<sup>14</sup> Rapid increases in plasma fibrinogen concentrations after surgery in both the fibrinogen group and the control groups is caused by up-regulation of hepatic fibrinogen synthesis resulting from the acute phase response to tissue injury.<sup>29-31</sup>

Table 3. Secondary Transfusion Outcomes<sup>a</sup>

	Fibrinogen (n = 58)	Control (n = 59)
<b>Patients who received transfusion between intervention and chest closure, No. (%)</b>		
Red blood cells	0	3 (5)
Fresh-frozen plasma	0	1 (2)
Platelets	2 (3)	2 (3)
Any transfusion	2 (3)	4 (7)
<b>Patients transfused between intervention and 24 h, thereafter, No. (%)</b>		
Red blood cells	10 (17)	20 (33)
Fresh-frozen plasma	9 (15)	13 (22)
Platelets	9 (15)	13 (22)
Any transfusion	20 (33)	23 (38)
<b>Transfusion units between intervention and 24 h thereafter, median (IQR)</b>		
Red blood cell transfusion units	0 (0-1)	0 (0-4)
Fresh-frozen plasma transfusion units	0 (0-2)	0 (0-4)
Platelets transfusion units	0 (0-1)	0 (0-1)
Any transfusion units	0 (0-2)	0 (0-8)

<sup>a</sup> Time point "intervention" is defined as moment of infusion of study medication.

Table 4. Clinical Adverse Events Within 30 Days

	No. of Events <sup>a</sup>	
	Fibrinogen (n = 60)	Control (n = 60)
In-hospital mortality	2	0
Stroke	4	1
Transient ischemic attack	0	1
Myocardial infarction	3	1
Renal insufficiency or failure	3	2
Thromboembolism	0	0
Allergic reaction	0	0
Infections	3	2
Rethoracotomy (≤5 d)	4	5

<sup>a</sup> Patients could have experienced more than 1 event.

Plasma fibrinogen levels in the control group fell and then rose on their own in our study as is shown in Figure 2. Fibrinogen levels were higher in the infusion group only within the first 24 hours after surgery suggesting that any effect of the fibrinogen infusion itself on controlling bleeding should have been experienced in that period.

Based on prior studies that were relied on to determine the sample size, the observed bleeding in the current study was less than expected. For the primary outcome, this can be explained by the fact that the period between the infusion of study medication and the closure of chest (the time frame for determination of primary blood loss), was relatively short. The exploratory analysis of the total blood loss in 24 hours assessed by mixed-model regression showed a 120 mL difference between the fibrinogen and control groups. Although statistically significant, this difference was less than the minimal clinically important difference anticipated for this study and, therefore, not clinically relevant. Furthermore, the significant *P* value for the exploratory secondary end point might be due to multiple comparisons and should be interpreted with caution.

In general, this study had a high-risk patient population; patients were older, had a high incidence of diabetes mellitus, and had long durations of cardiopulmonary bypass, which might explain the large number of adverse events.<sup>32</sup> All patients in this study who experienced a stroke had a postintervention plasma fibrinogen concentration lower than the targeted 2.5 g/L. Approximately 45% of perioperative strokes occur within the first 24 hours after surgery and are often related to the physical handling of the heart and aorta resulting in dislodgement of atherosclerotic debris.<sup>33-35</sup> These

effects might explain strokes observed in the first 24 hours of this study. Routine perioperative screening by echocardiography (transesophageal echocardiography [TEE], modified TEE, and epi-aortic scanning) for the severity of aortic atherosclerosis was not performed in this study.<sup>36</sup> This study was not powered to assess statistically significant differences in adverse events between the treatment and control group. In addition, the effect of the combined use of fibrinogen concentrate with other prothrombotic agents is unknown and may be harmful.

The study has some limitations. First, despite randomization, inequalities between the 2 groups occurred. Therefore, a multivariable analysis was performed to reduce the effect of these baseline differences on our results. Second, fibrinogen concentrate infusion was given postoperatively by the clinicians providing care for the study patients in 1 patient (2%) in the fibrinogen group and 6 patients (10%) in the control group, confounding the effect of fibrinogen concentrate on overall outcomes (eTable 2 in the Supplement). Third, this study was a single-center study and may not generalize to other care settings, although being a single-center coordinated study reduced the potential for variability in the study and adherence to the study protocol.

## Conclusions

Among patients with intraoperative bleeding during high-risk cardiac surgery, administration of fibrinogen concentrate, compared with placebo, resulted in no significant difference in the amount of intraoperative blood loss

### ARTICLE INFORMATION

**Author Contributions:** Drs Bilecen and Dr de Groot had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis.

**Concept and design:** Bilecen, Kalkman, Spanjersberg, Brandon Bravo Bruinsma, Moons, Nierich.

**Acquisition, analysis, or interpretation of data:** All authors.

**Drafting of the manuscript:** Bilecen, de Groot, Kalkman, Brandon Bravo Bruinsma, Moons, Nierich.

**Critical revision of the manuscript for important intellectual content:** All authors.

**Statistical analysis:** Bilecen, de Groot, Kalkman, Moons.

**Administrative, technical, or material support:** Bilecen, Spanjersberg, Brandon Bravo Bruinsma.

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