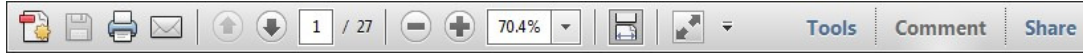
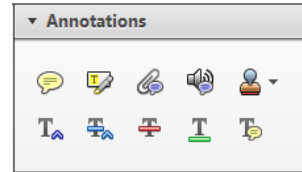


Once you have Acrobat Reader open on your computer, click on the [Comment](#) tab at the right of the toolbar:



This will open up a panel down the right side of the document. The majority of tools you will use for annotating your proof will be in the [Annotations](#) section, pictured opposite. We've picked out some of these tools below:



### 1. Replace (Ins) Tool – for replacing text.

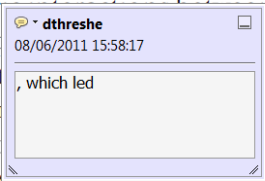


Strikes a line through text and opens up a text box where replacement text can be entered.

#### How to use it

- Highlight a word or sentence.
- Click on the [Replace \(Ins\)](#) icon in the Annotations section.
- Type the replacement text into the blue box that appears.

standard framework for the analysis of microeconomic activity. Nevertheless, it also led to the development of a number of strategic approaches. The number of competitors in an industry is that the structure of the industry is a key component. At the firm level, are externalities important? (Mankiw henceforth) we open the 'black b



### 2. Strikethrough (Del) Tool – for deleting text.



Strikes a red line through text that is to be deleted.

#### How to use it

- Highlight a word or sentence.
- Click on the [Strikethrough \(Del\)](#) icon in the Annotations section.

there is no room for extra profits as mark-ups are zero and the number of firms (net) values are not determined by market structure. Blanchard ~~and Kiyotaki~~ (1987), perfect competition in general equilibrium. The effects of aggregate demand and supply shocks in the classical framework assuming monopolistic competition are an exogenous number of firms

### 3. Add note to text Tool – for highlighting a section to be changed to bold or italic.



Highlights text in yellow and opens up a text box where comments can be entered.

#### How to use it

- Highlight the relevant section of text.
- Click on the [Add note to text](#) icon in the Annotations section.
- Type instruction on what should be changed regarding the text into the yellow box that appears.

dynamic responses of mark-ups are consistent with the VAR evidence

sation of the industry. The number of competitors and the impact of demand shocks on the industry are also with the demand-



### 4. Add sticky note Tool – for making notes at specific points in the text.

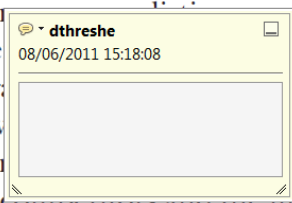


Marks a point in the proof where a comment needs to be highlighted.

#### How to use it

- Click on the [Add sticky note](#) icon in the Annotations section.
- Click at the point in the proof where the comment should be inserted.
- Type the comment into the yellow box that appears.

and supply shocks. Most of the time, the number of firms in an industry is determined by market structure. The effects of aggregate demand and supply shocks in the classical framework assuming monopolistic competition are an exogenous number of firms



5. **Attach File** Tool – for inserting large amounts of text or replacement figures.

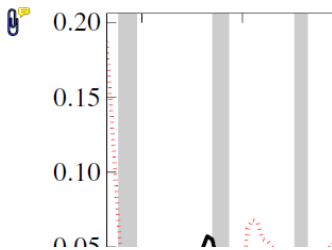


Inserts an icon linking to the attached file in the appropriate place in the text.

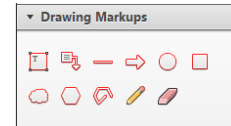
How to use it

- Click on the **Attach File** icon in the Annotations section.
- Click on the proof to where you'd like the attached file to be linked.
- Select the file to be attached from your computer or network.
- Select the colour and type of icon that will appear in the proof. Click OK.

END

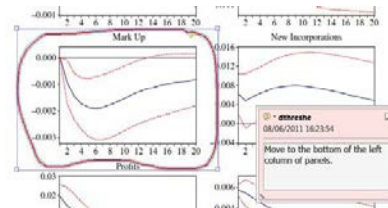


6. **Drawing Markups** Tools – for drawing shapes, lines and freeform annotations on proofs and commenting on these marks. Allows shapes, lines and freeform annotations to be drawn on proofs and for comment to be made on these marks.



How to use it

- Click on one of the shapes in the Drawing Markups section.
- Click on the proof at the relevant point and draw the selected shape with the cursor.
- To add a comment to the drawn shape, move the cursor over the shape until an arrowhead appears.
- Double click on the shape and type any text in the red box that appears.



# Desmopressin after cardiac surgery in bleeding patients. A multicenter randomized trial

E. Bignami<sup>1</sup>, M. Cattaneo<sup>2</sup>, G. Crescenzi<sup>1</sup>, M. Ranucci<sup>3</sup>, F. Guarracino<sup>4</sup>, C. Cariello<sup>4</sup>, R. Baldassarri<sup>4</sup>,  
G. Isgrò<sup>3</sup>, E. Baryshnikova<sup>3</sup>, G. Fano<sup>1</sup>, A. Franco<sup>1</sup>, C. Gerli<sup>1</sup>, M. Crivellari<sup>1</sup>, A. Zangrillo<sup>1,5</sup> and G. Landoni<sup>1,5</sup>

<sup>1</sup>Department of Anesthesia and Intensive Care, IRCCS San Raffaele Scientific Institute, Milan, Italy

<sup>2</sup>Dipartimento di Scienze della Salute, Ospedale San Paolo, Università degli Studi di Milano, Milan, Italy

<sup>3</sup>Department of Anesthesia and Intensive Care, IRCCS Policlinico San Donato, Milan, Italy

<sup>4</sup>Department of Anesthesia and Intensive Care, University Hospital of Pisa, Pisa, Italy

<sup>5</sup>Vita-Salute San Raffaele University, Milan, Italy

## Correspondence

G. Landoni, Department of Anesthesia and Intensive Care, IRCCS San Raffaele Scientific Institute, Vita-Salute San Raffaele University, Via Olgettina 58-60, 20132 Milan, Italy

**E-mail:** landoni.giovanni@hsr.it

## Conflict of interest

The authors have no conflict of interest to declare.

## Funding

The manuscript was supported by departmental funds only.

## Trial registration number

ClinicalTrials.gov no. NCT00337766. <https://www.clinicaltrials.gov/ct2/show/NCT00337766?term=NCT00337766&rank=1>.

Submitted 5 April 2016; accepted 6 April 2016; submission 12 January 2016.

## Citation

Bignami E, Cattaneo M, Crescenzi G, Ranucci M, Guarracino F, Cariello C, Baldassarri R, Isgrò G, Baryshnikova E, Fano G, Franco A, Gerli C, Crivellari M, Zangrillo A, Landoni G. Desmopressin after cardiac surgery in bleeding patients. A multicenter randomized trial. *Acta Anaesthesiologica Scandinavica* 2016

doi: 10.1111/aas.12740

**Background:** Previous studies showed that desmopressin decreases post-operative blood loss in patients undergoing cardiac surgery. These studies were small and never studied the effect of desmopressin in patients with active bleeding. Objective of the study was to determine whether desmopressin reduces red blood cells transfusion requirements in patients with active bleeding after cardiac surgery who had been pre-treated with tranexamic acid.


**Methods:** This multicenter, randomized, double-blind, placebo-controlled, parallel-group study randomized elective patients with bleeding after cardiac surgery despite pre-treatment with tranexamic acid, to receive placebo (saline solution) or a single administration of desmopressin (0.3 µg/kg in saline solution). The primary endpoint was the number of patients requiring red blood cells transfusion after randomization and during hospital stay. Secondary end points were: blood loss from chest tubes during the first 24 h after study drug administration, hours of mechanical ventilation, intensive care unit stay, and in-hospital mortality.

**Results:** The study was interrupted after inclusion of 67% of the planned patients for futility. The number of patients requiring red blood cells transfusion after randomization was 37/68 (54%) in desmopressin group and 33/67 (49%) in placebo group ( $P = 0.34$ ) with no difference in blood loss: 575 (interquartile 422–770) ml in desmopressin group and 590 (476–1013) ml in placebo group ( $P = 0.42$ ), mechanical ventilation, intensive care unit stay or mortality.

**Conclusions:** This multicenter randomized trial demonstrated that, in patients pre-treated with tranexamic acid, desmopressin should not be expected to improve treatment of patients who experience bleeding after cardiac surgery.

## Editorial comment

Desmopressin is sometimes administered to perioperative patients in the hope of reducing perioperative bleeding. The findings from this clinical trial indicate that in patients treated with tranexamic acid before cardiac surgery who then have perioperative bleeding, the addition of desmopressin treatment probably will not reduce bleeding.

Dispatch: 18.4.16	CE: Madhumitha R
No. of pages: 9	PE: Sharmila
WILEY	
12740	Manuscript No.
AAS	Journal Code
	

1 Peri-operative bleeding and the need for blood  
2 transfusions are correlated with increased mor-  
3 bidity, mortality, and costs.<sup>1-4</sup> In cardiac sur-  
4 gery, patients often require transfusions of  
5 blood products and a surgical cause cannot be  
6 identified in many patients. Among non-surgical  
7 factors, hemostatic abnormalities are of para-  
8 mount importance.

9 Pharmacological options to control peri-oper-  
10 ative bleeding consist of topical agents, antifibri-  
11 nolytic drugs, and drugs that improve primary  
12 and/or secondary hemostasis. Among the latter  
13 drugs, desmopressin (1-deamino-8-D-arginine  
14 vasopressin, DDAVP) has been extensively  
15 studied in several types of surgical interven-  
16 tion,<sup>5</sup> but mostly in patients undergoing cardiac  
17 surgery with extracorporeal circulation.<sup>6,7</sup>  
18 Desmopressin stimulate increases in plasma  
19 concentrations of factor VIII and von Willebrand  
20 factor and exerts other, less well-characterized  
21 pro-hemostatic effects.<sup>8-10</sup>

22 Several randomized controlled trials  
23 (RCTs)<sup>11,12</sup> and a meta-analysis of RCTs<sup>5</sup> sug-  
24 gested that prophylactic desmopressin reduces  
25 bleeding in surgical patients with a clinical  
26 effect of approximately 3-4 h.<sup>13</sup> Existing guide-  
27 lines agree with the potential hemostatic proper-  
28 ties of desmopressin, but do not give strong  
29 recommendations.<sup>14,15</sup> Notably, all the RCTs  
30 performed so far have a small sample size, none  
31 of them included patients with active post-  
32 operative bleeding, and only one of them was  
33 multicenter.

34 With the hypothesis that desmopressin might  
35 reduce the transfusion rate of red blood cells  
36 (RBC) in patients with excessive bleeding after  
37 cardiac surgery, we performed a multicenter  
38 RCT comparing a single dose infusion of desmo-  
39 pressin vs. placebo. We also investigated in-hos-  
40 pital mortality, blood loss/24 h, hours of  
41 mechanical ventilation, duration of intensive  
42 care unit, and hospital stay.

## 43 Methods

44 We performed a multicenter, randomized, dou-  
45 ble-blind, placebo-controlled, parallel-group  
46 trial to determine if desmopressin could reduce  
47 the need for packed RBC transfusion after car-  
48 diac surgery in patients with excessive bleeding

in the operating room or in the cardiac  
post-operative intensive care unit (ICU). The  
drug desmopressin is used in a way that is dif-  
ferent from that described in the Food and Drug  
Administration-approved drug label ('off-label'  
use).

Three teaching centers participated to the  
study, all in Italy: IRCCS San Raffaele Scientific  
Institute, Milan; IRCCS Policlinico San Donato,  
Milan; and University Hospital of Pisa, Pisa.

The study was conceived in accordance with  
the Declaration of Helsinki and its amendments.  
The study protocol was approved by the Ethical  
Committees of the three teaching centers  
involved (Coordinating Center: Ospedale San  
Raffaele Ethical Committee, IRCCS San Raffaele  
Scientific Institute, via Olgettina 60, 20132  
Milan; protocol n. DS/URC/ER/mm 151/DG;  
date of approval: March 13th, 2006), registered  
on ClinicalTrials.gov with the identifier  
NCT00337766 and the first patient was random-  
ized on June 2006.

All patients aged 18 years or more undergoing  
elective cardiac surgery were asked to sign the  
informed consent before surgery and if they met  
the inclusion criteria they were randomized to  
receive either desmopressin or placebo, in the  
operating room or in the post-operative cardiac  
ICU.

## 49 Inclusion/exclusion criteria

The inclusion criterion in the operating room  
was diffuse bleeding without an identifiable  
surgical source from the surgical site  $\geq 15$  min  
after correct antagonization of heparin with pro-  
tamine,<sup>12</sup> likely to require management with  
transfusion of platelets concentrates, fresh frozen  
plasma, and/or RBC. This was decided jointly  
by surgeons and anesthesiologists. The inclu-  
sion criterion in the ICU was excessive bleeding  
from chest tubes defined as 200 ml/h in 30 min  
or 2 ml/kg/h for at least 2 h.<sup>16</sup> Exclusion criteria  
were lack of informed consent, and recent  
( $< 7$  days) or ongoing acute myocardial infar-  
ction.

## 50 Randomization and experimental procedure

A computer-generated randomization sequence  
stratified by center with blocks of 20 was used.

1 Treatment allocation (1 : 1) prepared by an  
2 independent operator not otherwise involved in  
3 the trial was concealed by opaque, sealed envel-  
4 opes that were sequentially numbered. Patients,  
5 managing physicians, and nurses were blinded  
6 to treatment assignment for the whole duration  
7 of the study. Desmopressin and placebo were  
8 prepared in a separate room, as colorless fluids  
9 in unlabeled bottles, by personnel who was not  
10 involved in patients' management and data col-  
11 lection. Desmopressin (Kedrion, Barga, Lucca,  
12 Italy) or an equivolume saline solution was  
13 administered intravenously over 20 min at a  
14 dose of 0.3 µg/kg diluted in a 50 ml saline solu-  
15 tion. This dose was chosen because it was the  
16 most frequently used (35 out of 41) in previous  
17 RCTs performed in the peri-operative period.<sup>5</sup>  
18 Registration of blood loss was started after com-  
19 pleting study drug administration. Data were  
20 collected by trained personnel who did not par-  
21 ticipate in patients' management and was  
22 blinded to the assigned treatment. No protocol  
23 violations were noted.

### 24 25 26 **Clinical management**

27 Enrolled patients were managed according to  
28 standardized institutional practice for cardiac  
29 surgery. This was a pragmatic trial and all  
30 patients received the standard treatment accord-  
31 ing to the anesthesiologist and the physician in  
32 charge of the ICU. In patients on treatment with  
33 double antiplatelet therapy, aspirin was main-  
34 tained throughout the surgical procedure, while  
35 drugs inhibiting the platelet P2Y<sub>12</sub> receptor  
36 were discontinued at least 5 days before sur-  
37 gery. Patient monitoring included invasive  
38 radial artery blood pressure measurement, con-  
39 tinuous electrocardiographic leads II and V5  
40 monitoring with ST-segment monitoring, pulse  
41 oximetry, central venous pressure, capnometry,  
42 urine output and trans-esophageal echocardiog-  
43 raphy. All patients received an intravenous slow  
44 bolus of 1 g tranexamic acid before surgery,  
45 which was followed by a continuous infusion of  
46 400 mg/h during the surgical intervention in  
47 one institution (San Raffaele Scientific Insti-  
48 tute), or by a second intravenous 1 g bolus after  
49 the administration of protamine in the remain-  
50 ing two institutions. A further dose of 500 mg  
51 tranexamic acid was allowed after cardio

pulmonary bypass (CPB). Aprotinin was never  
administered.

### **CPB management**

Surgery was conducted either with CPB or off  
pump. Cardio pulmonary bypass was conducted  
mostly at moderate hypothermia (32–34°C). In  
CPB procedures, myocardial protection during  
aortic cross-clamping was obtained by antegrade  
and/or retrograde cold blood cardioplegia or  
antegrade crystalloid cardioplegia according to  
the local standard. Heparin was administered as  
intravenous bolus of 300 IU/kg to patients under-  
going CPB, or 150 IU/kg to patients undergoing  
off pump procedures. Activated clotting time was  
maintained longer than 480 s during CPB and  
longer than 300 s during off pump procedures.  
Upon achievement of normothermia, extracorp-  
oreal circulation was discontinued and heparin  
was neutralized with protamine sulfate (1 mg per  
100 IU heparin).

### **Intensive care management**

Patients were transferred to the ICU, sedated  
with propofol 2 mg/kg/h for 4 h, and weaned  
from the ventilator as soon as they were hemo-  
dynamically stable with no major bleeding, nor-  
mothermic, and achieved an adequate level of  
consciousness and analgesia. Intravenous mor-  
phine was administered to all patients for post-  
operative pain relief. In case of bleeding, the  
surgeon was summoned to decide if surgical  
revision was suitable. Criteria for ICU discharge  
to post-intensive cardiac surgery ward were res-  
piratory and hemodynamic stability and no  
active bleeding from chest tubes.

### **Bleeding and transfusion management**

The amount of blood loss from chest tubes was  
recorded hourly for 48 h after surgery or until  
chest tubes removal, whichever came first. The  
number of transfused platelet concentrates, fresh  
frozen plasma, and RBC were recorded. Transfu-  
sions were administered following a pre-  
defined protocol: if hemoglobin was < 8 g/dl,  
one RBC was transfused. In case of bleeding  
from chest tubes, if prothrombin time (INR)  
was ≤ 1.39, no fresh frozen plasma was infused,



if platelets count was  $> 100 \times 10^9/l$ , no platelets concentrates were infused. In the operating room, advanced patient's age or hemodynamic instability were factors that allowed a lower threshold for RBC transfusion, according to the attending anesthesiologist.

### Data collection

We collected data on need of re-thoracotomy due to excessive bleeding, length of mechanical ventilation and of ICU stay. All randomized patients were followed up till hospital discharge.

Laboratory-based hematological tests, including complete blood cells count, prothrombin time ratio, PTT ratio, and biochemistry tests, including serum creatinine, troponin I, transaminase, and plasma sodium concentration were done before and after surgery, on arrival in ICU and on the first post-operative day. The anticoagulant effect of heparin was monitored by kaolin-activated clotting time before and after CPB.

### Endpoints

The pre-specified main outcome measure was the number of patients requiring RBC transfusion after randomization and during hospital stay. Secondary end points were: blood loss from chest tubes during the first 24 h after study drug administration, hours of mechanical ventilation, ICU stay, and in-hospital mortality.

### Sample size calculation

On the basis of previous data<sup>17,18</sup> investigating post-operative RBC transfusion in patients at high risk of bleeding after cardiac surgery and on our experience, we anticipated a 65% frequency of transfusion of RBC in the standard treatment group and a reduction to 45% after treatment with desmopressin (we tested the hypothesis that desmopressin administration would reduce the need of RBC transfusion when compared to placebo with an absolute risk reduction of 20% and a relative risk reduction of 30%). Sample size calculations, based on a two-sided alpha error of 0.05 and 80% power, indicated that a sample size of 100 patients per group was necessary.

### Ad interim analysis and study interruption

We experienced a low enrollment rate due to organizing problems and lack of funding and we decided to perform an unplanned ad interim analysis after enrolling 67% of the patients. As showed in supplemental material (Appendix S1, Figs S1 and S2) the trial was considered to have reached its goal of testing the null hypothesis with adequate statistical precision and was stopped for futility.

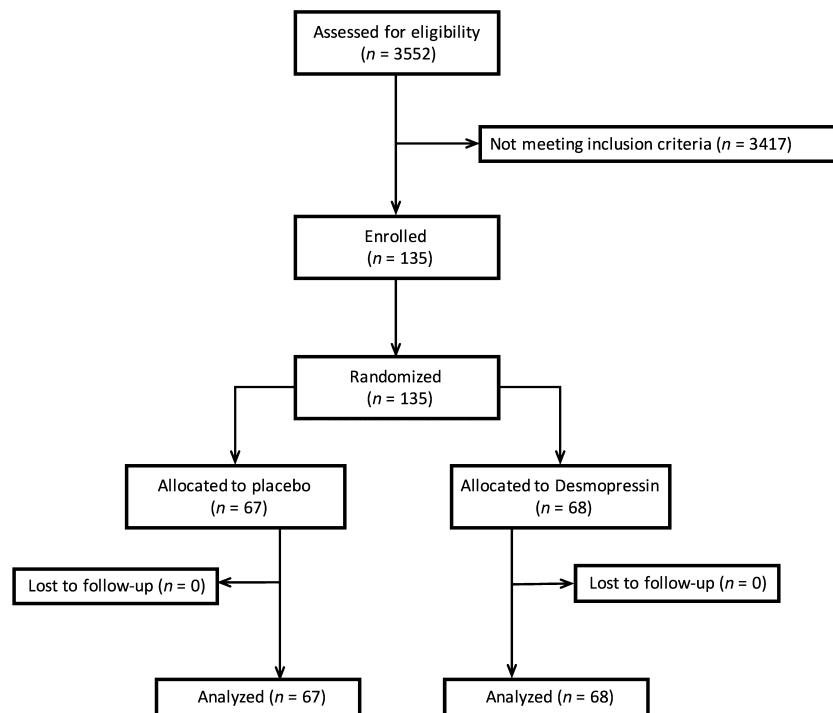
### Statistical analyses

All data were analyzed according to the intention-to-treat principle. Data were stored electronically and analyzed by use of SAS software, version 9 (SAS Institute). All data analysis was carried out according to a pre-established analysis plan and results are presented as mean  $\pm$  standard deviation, median (interquartile range), or number (percentage). Dichotomous data were compared using two-tailed Chi-square test with the Yates correction or Fisher's exact test when appropriate; 95% confidence interval estimation for the differences between independent proportions was performed with methods based on the Wilson score. Continuous measures were compared by analysis of variance (ANOVA) or the Mann-Whitney *U*-test when appropriate; 95% confidence interval estimate for the mean/median difference was performed. Two-sided significance tests were used throughout our data analysis.

### Results

During the study period, 3552 patients signed the written informed consent. A total of 135 (3.8%) patients had excessive bleeding after cardiac surgery and were randomized to desmopressin ( $n = 68$ ) or placebo ( $n = 67$ ) (Fig. 1). The study was interrupted for futility at an ad interim analysis performed after enrolling 67% of the planned patients. All 135 included patients completed their hospital follow-up.

The two groups of patients had similar characteristics at baseline (Table 1). Mean age was 63 years, 100 (74%) were males and 54 (40%) were on treatment with antiplatelet drugs. Seventy-three patients (54%) were randomized



**Fig. 1.** Assessment, randomization and follow-up of study patients.

in the operating theater and 62 (46%) in the ICU after a median of 120 (60–180) min after arrival to ICU.

All patients received a single intravenous administration of placebo or desmopressin 0.3 µg/kg, without protocol deviations.

The number of patients receiving RBC transfusions after randomization was similar in the desmopressin (37/68, 54%) and in the placebo group (33/67, 49%) ( $P = 0.34$ ) (Table 2).

No statistically significant differences were observed between the two treatment groups in terms of hours of mechanical ventilation, days in ICU and other outcomes (Table 2). Five patients (7.4%) in the desmopressin group and 4 (6.0%) in the placebo group died ( $P = 0.99$ ).

Neither differences in bleeding nor RBC transfusion was observed in subanalyses of patients randomized in the theater (Table S1) and those randomized in the ICU (Table S2).

## Discussion

In this multicenter, double-blind, placebo-controlled RCT, we found that desmopressin was not effective in reducing post-operative blood loss and RBC transfusion requirements in

patients undergoing cardiac surgery who experienced excessive bleeding after successful heparin neutralization despite pre-treatment with tranexamic acid. Moreover, no statistically significant differences in mortality rates or in any other outcomes were observed, even if the power for these analyses was low. Transfusion of RBC did not differ significantly also in pre-defined subgroups. The study was interrupted for futility after enrolling 67% of the planned patients.

Previous studies evaluating the blood saving effects of desmopressin in patients undergoing cardiac surgery gave conflicting results. A meta-analysis of 17 studies<sup>17</sup> showed that desmopressin decreased post-operative blood loss by 34% in those trials in which the 24 h blood loss in the placebo-treated patients was in the upper third of distribution, suggesting that the drug may be clinically efficacious only in patients at risk of excessive post-operative bleeding.<sup>17</sup> The data of this meta-analysis were substantially confirmed in a later prospective, double-blind, placebo-controlled trial, in which the effects of desmopressin were compared to those of placebo in patients identified as being at high risk of post-operative bleeding with the point-of-care

**Table 1** Baseline characteristics according to randomized group assignment (desmopressin vs. placebo).

Variable	Desmopressin (68 patients)	Placebo (67 patients)
Age, years	64 ± 13.3	62 ± 13.2
No. of patients aged > 75 years	15 (23%)	13 (20%)
Women	18 (27%)	17 (25%)
Weight, kg	73 ± 13.5	73 ± 13.8
Height, cm	169 ± 8.4	170 ± 8.1
Cirrhosis, no.	2 (3%)	1 (1.5%)
On antiplatelet drugs, no.	25 (38%)	29 (43%)
Acetylsalicylic acid, no.	21 (31%)	16 (24%)
Ticlopidine, no.	4 (5.9%)	5 (7.5%)
Clopidogrel, no.	8 (12%)	9 (13%)
Tirofiban, no.	1 (1.5%)	1 (1.5%)
Congenital defect of primary hemostasis, no.	1 (1.5%)	1 (1.5%)
Activated clotting time before surgery, seconds	147 ± 29.4	145 ± 24.4
Activated clotting time at end of CPB, seconds	140 ± 29.0	140 ± 20.7
Hemoglobin preoperative, g/dl	13.3 ± 1.85	13.4 ± 1.89
Hematocrit preoperative, %	39.3 ± 5.19	39.6 ± 5.15
Platelet count preoperative, × 10 <sup>3</sup> /mm <sup>3</sup>	204 ± 73.4	204 ± 60.5
PT ratio preoperative	1.2 ± 0.31	1.2 ± 0.53
PTT ratio preoperative	1.1 ± 0.25	1.2 ± 0.39
Serum creatinine preoperative, mg/dl	0.90 (0.79–1.08)	0.96 (0.82–1.1)
No. of patients with preoperative creatinine > 1.5 mg/dl	7 (11%)	5 (7.6%)
Randomization in the operating theater, no.	37 (54%)	36 (54%)
Minutes between CPB weaning and randomization in the theater	60 (30–80)	50 (40–79.8)
Randomization in the ICU, no.	31 (46%)	31 (46%)
Minutes between ICU admission and randomization in the ICU	120 (60–157.5)	145 (60–240)
Supplemental dose of tranexamic acid, no.	35 (52%)	34 (51%)
No. of patients transfused with any blood product before randomization	31 (46%)	34 (51%)
No. of patients transfused with RBC before randomization	24 (35%)	28 (42%)
No. of RBC units transfused/patient before randomization	2 (1.75–3)	2 (1–3)
No. of patients transfused with FFP pre-randomization	20 (29%)	21 (31%)
No. of FFP units transfused/patient before randomization	2 (1.75–3.25)	2 (2–3)
No. of patients transfused with PLT pre-randomization	10 (15%)	9 (13%)
No. of PLT units transfused/patient before randomization	1 (1–1)	1 (1–1)

No., number; CPB, cardiopulmonary bypass; PT, prothrombin time; INR, international normalized ratio; PTT, partial thromboplastin time; Na, sodium; ICU, intensive care unit; RBC, red blood cell; FFP, fresh frozen plasma; PLT, platelets.

test hemoSTATUS.<sup>12</sup> The trial showed that the average 24 h blood loss in desmopressin-treated, high-risk patients was 39% lower than that in placebo-treated high-risk patients and was comparable to that of low risk, untreated patients. The most important difference between our study and previous studies is that as part of modern cardiac surgery<sup>19,20</sup> we used tranexamic acid in all patients included in the present trial. Interestingly, 1000 ml of blood loss in the first post-operative 24 h seems to be the cut-off value that identifies patients who may potentially benefit from this treatment.<sup>19</sup> Therefore, we speculate that, when an adequate prophylaxis

with antifibrinolytic agents is administered to patients, excessive post-operative bleeding is not severe enough to benefit from the pro-hemostatic effect of desmopressin.

In a recent meta-analysis of 38 randomized placebo-controlled trials (2488 patients) in cardiac and non-cardiac surgery, Crescenzi et al. showed that desmopressin was associated with reduced requirements of blood product transfusion [standardized mean difference = -0.29 (-0.52 to -0.06) units per patient; *P* = 0.01], and with a non-statistically significant increase in thromboembolic adverse events (57/1002 = 5.7% in the desmopressin group vs. 45/979 = 4.6% in the



**Table 2** Post-randomization data according to randomized group assignment (desmopressin vs. placebo).

Variable	Desmopressin (68 patients)	Placebo (67 patients)	P value
No. of patients transfused with RBC post-randomization	37 (54%)	33 (49%)	0.34
No. of RBC units transfused/patient post-randomization	2 (1–4)	2 (1–3)	0.74
No. of patients transfused with FFP post-randomization	24 (35%)	25 (37%)	0.62
No. of FFP units transfused/patient post-randomization	2.5 (2–4)	3 (2–3)	0.77
No. of patients transfused with PLT post-randomization	9 (13%)	10 (15%)	0.62
No. of PLT units transfused/patient post-randomization	1 (1–3)	1 (1–2)	0.90
No. of patients transfused with any blood product post-randomization	40 (59%)	35 (52%)	0.44
Chest tube blood loss in the first 12 h after study drug administration, ml	400 (267.5–700)	430 (325–800)	0.34
Chest tube blood loss in the first 24 h after study drug administration, ml	575 (422.5–770)	590 (476.25–1013.75)	0.42
Bleeding at drainage removal, ml	850 (600–1390)	1040 (565–1457.5)	0.69
Hemoglobin at ICU admission, g/dl	11.1 ± 1.74	11.1 ± 1.68	0.67
Hemoglobin in the 1st morning, g/dl	10.8 ± 1.36	11.1 ± 1.46	0.16
Hemoglobin in the 2nd morning, g/dl	10.6 ± 1.41	10.7 ± 1.23	0.78
Hematocrit at ICU admission, %	33.1 ± 4.70	33.1 ± 4.83	0.98
Hematocrit in the 1st morning, %	31.8 ± 4.08	32.7 ± 4.04	0.23
Hematocrit in the 2nd morning, %	31.0 ± 4.14	31.3 ± 3.90	0.68
Platelet count at ICU admission, ×10 <sup>3</sup> /mm <sup>3</sup>	136 ± 69.8	128 ± 47.6	0.47
Platelet count in the 1st morning, ×10 <sup>3</sup> /mm <sup>3</sup>	136 ± 62.4	140 ± 58.0	0.74
Platelet count in the 2nd morning, ×10 <sup>3</sup> /mm <sup>3</sup>	135 ± 68.1	128 ± 55.0	0.63
PT ratio at ICU admission	1.4 ± 0.32	1.4 ± 0.28	0.65
PT ratio in the 1st morning	1.3 ± 0.21	1.2 ± 0.18	0.61
PT ratio in the 2nd morning	1.4 ± 0.48	1.3 ± 0.25	0.94
PTT ratio at ICU admission	1.1 ± 0.27	1.2 ± 0.34	0.85
PTT ratio in the 1st morning	1.1 ± 0.12	1.0 ± 0.10	0.85
PTT ratio in the 2nd morning	1.1 ± 0.19	1.1 ± 0.15	0.66
Serum creatinine at ICU admission, mg/dl	0.97 (0.82–1.09)	0.89 (0.75–1.09)	0.23
Serum creatinine in the 1st morning, mg/dl	1.15 (0.95–1.4)	1.22 (0.89–1.56)	0.94
Serum creatinine peak level in the ICU, mg/dl	1.23 (1.02–1.71)	1.14 (0.95–1.57)	0.37
Serum troponin I at ICU admission, ng/l	4.1 (2.1–8.4)	5.1 (2.1–8.9)	0.73
Serum troponin I in the 1st morning, ng/l	6.8 (2.6–11.3)	5.8 (3.3–11.1)	0.97
Serum troponin I peak level, ng/l	8.4 (3.4–11.5)	7.5 (3.9–13.6)	0.70
Hypotension during study drug administration, no.	12 (18%)	4 (6.0%)	0.11
Hypotension requiring intervention, no.	3 (4.4%)	2 (3.0%)	0.90
Surgical revision, no.	8 (12%)	12 (18%)	0.39
Surgical source of bleeding in re-operated patients, no.	5/8 (62%)	5/12 (42%)	0.36
Mechanical ventilation, h	17 (11–34)	16 (10.75–31)	0.92
ICU stay, days	2 (1–4)	2 (1–4)	0.63
Thrombosis, no.	3 (4.4%)	2 (3.0%)	0.51
Hospital mortality, no.	5 (7.4%)	4 (6.0%)	0.99

No., number; ICU, intensive care unit; RBC, red blood cell; FFP, fresh frozen plasma; PLT, platelets; PT, prothrombin time; INR, international normalized ratio; PTT, partial thromboplastin time.

placebo group;  $P = 0.3$ ).<sup>6</sup> Our study will also help to modify the message of the existing guidelines<sup>14,15</sup> for the peri-operative management of desmopressin. At the moment, they are both suggesting potential benefits in reducing blood loss in the presence of coagulopathy conditions, but they are based on small single-center trials or meta-analysis<sup>21</sup> performed in settings that did not routinely use tranexamic acid. In this

multicenter randomized trial desmopressin did not translate into a reduced rate of patients requiring RBC transfusion among those with active bleeding after cardiac surgery who had been pre-treated with tranexamic acid.

Importantly, it is necessary to investigate further how to improve outcomes of patients with bleeding after surgery, which has a high mortality rate, as confirmed by the results of our

study: 6.7% in the patients included in this study vs. 2.2–3.4% published<sup>22,23</sup> mortality reported in the overall population undergoing cardiac surgery in our centers.

This trial was randomized and double-blinded in design with allocation concealment, thus reducing the risk of selection bias. It focused on patient-centered, objectively verifiable and clinically relevant outcomes, thus reducing ascertainment bias. The intervention had biological plausibility and was supported by a series of single-center studies with promising results and by meta-analyses, thus justifying the initial trial hypothesis. Our results likely carry external validity because patients were recruited in three different hospitals with the use of pragmatic inclusion criteria and few exclusion criteria. They are also likely to be highly reproducible, as the trial protocol was simple, with routine practice maintained throughout, except for desmopressin or placebo infusion.

A limitation of our study is that it was interrupted prematurely, after the enrollment of 135 of the planned 200 patients; even if this caused a reduction in power, the case for the futility of the intervention was clear. Despite its premature interruption, this is the largest multicenter RCT ever performed with desmopressin as a hemostatic agent in the peri-operative period and one of the few to study patients who were actively bleeding. This was a pragmatic trial and we did not change the routine of our centers to perform the protocol; at the same time we acknowledge that we did not collect some important data that could influence coagulability such as kind of surgery (e.g., off pump CABG); CPB duration, pH, calcium, and the number of patients operated under hypothermia or with normothermia. A possible limitation of our study is that the inclusion criteria in the theater might be considered less objective and relevant than the inclusion criteria in the ICU, but hemostasis is optimized in the operating rooms and other investigators randomized patients in this setting. Furthermore, results in the two groups (theater and ICU) were similar.

In conclusion, this multicenter randomized trial demonstrated that in patients pre-treated with tranexamic acid, desmopressin should not be expected to improve treatment of patients

who experience bleeding after cardiac surgery. Therefore, also considering its potential side effects, the routine use of desmopressin as a blood saving agent in this setting is not justified.

## Acknowledgements

The Authors gratefully acknowledge the technical contribution of Paola Zuppelli and Rosalba Lembo (San Raffaele Scientific Institute). None received compensation for their contributions.

## References

1. Ranucci M, Bozzetti G, Ditta A, Cotza M, Carboni G, Ballotta A. Surgical reexploration after cardiac operations: why a worse outcome? *Ann Thorac Surg* 2008; 86: 1557–62.
2. Koch CG, Li L, Duncan AI, Mihaljevic T, Cosgrove DM, Loop FD, Starr NJ, Blackstone EH. Morbidity and mortality risk associated with red blood cell and blood-component transfusion in isolated coronary artery bypass grafting. *Crit Care Med* 2006; 34: 1608–16.
3. Koch CG, Li L, Duncan AI, Mihaljevic T, Loop FD, Starr NJ, Blackstone EH. Transfusion in coronary artery bypass grafting is associated with reduced long-term survival. *Ann Thorac Surg* 2006; 81: 1650–7.
4. Leal-Noval SR, Rincón-Ferrari MD, García-Curiel A, Herruzo-Avilés A, Camacho-Laraña P, Garnacho-Montero J, Amaya-Villar R. Transfusion of blood components and postoperative infection in patients undergoing cardiac surgery. *Chest* 2001; 119: 1461–8.
5. Crescenzi G, Landoni G, Biondi-Zoccai G, Pappalardo F, Nuzzi M, Bignami E, Fochi O, Maj G, Calabrò MG, Ranucci M, Zangrillo A. Desmopressin reduces transfusion needs after surgery: a meta-analysis of randomized clinical trials. *Anesthesiology* 2008; 109: 1063–76.
6. Levi M, Cromheecke ME, de Jonge E, Prins MH, de Mol BJ, Briët E, Büller HR. Pharmacological strategies to decrease excessive blood loss in cardiac surgery: a meta analysis of clinically relevant endpoints. *Lancet* 1999; 354: 1940–7.
7. Wademan BH, Galvin SD. Desmopressin for reducing postoperative blood loss and transfusion requirements following cardiac surgery in adults. *Interact Cardiovasc Thorac Surg* 2014; 18: 360–70.
8. Cattaneo M. The use of desmopressin in open-heart surgery. *Haemophilia* 2008; 14(Suppl. 1): 40–7.

9. Cattaneo M, Moia M, Delle Valle P, Castellana P, Mannucci PM. DDAVP shortens the prolonged bleeding times of patients with severe von Willebrand disease treated with cryoprecipitate. Evidence for a mechanism of action independent of released von Willebrand factor. *Blood* 1989; 74: 1972–5.
10. Colucci G, Stutz M, Rochat S, Conte T, Pavicic M, Reusser M, Giabbani E, Huynh A, Thürlemann C, Keller P, Alberio L. The effect of desmopressin on platelet function: a selective enhancement of procoagulant COAT platelets in patients with primary platelet function defects. *Blood* 2014; 123: 1905–16.
11. Salzman EW, Weinstein MJ, Weintraub RM, Ware JA, Thurer RL, Robertson L, Donovan A, Gaffney T, Bertele V, Troll J, Smith M, Chute LE. Treatment with desmopressin acetate to reduce blood loss after cardiac surgery. A double-blind randomized trial. *N Engl J Med* 1986; 314: 1402–6.
12. Despotis GJ, Levine V, Saleem R, Spitznagel E, Joist JH. Use of point-of-care test in identification of patients who can benefit from desmopressin during cardiac surgery: a randomised controlled trial. *Lancet* 1999; 354: 106–10.
13. Agersø H, Seiding Larsen L, Riis A, Lövgren U, Karlsson MO, Senderovitz T. Pharmacokinetics and renal excretion of desmopressin after intravenous administration to healthy subjects and renally impaired patients. *Br J Clin Pharmacol* 2004; 58: 352–8.
14. Kozek-Langenecker SA, Afshari A, Albaladejo P, Santullano CA, De Robertis E, Filipescu DC, Fries D, Görlinger K, Haas T, Imberger G, Jacob M, Lancé M, Llau J, Mallett S, Meier J, Rahe-Meyer N, Samama CM, Smith A, Solomon C, Van der Linden P, Wikkelsø AJ, Wouters P, Wyffels P. Management of severe perioperative bleeding: guidelines from the European Society of Anaesthesiology. *Eur J Anaesthesiol* 2013; 30: 270–382.
15. American Society of Anesthesiologists Task Force on Perioperative Blood Management. Practice guidelines for perioperative blood management: an updated report by the American Society of Anesthesiologists Task Force on Perioperative Blood Management. *Anesthesiology* 2015; 122: 241–75.
16. Gill R, Herbertson M, Vuylsteke A, Olsen PS, von Heymann C, Mythen M, Sellke F, Booth F, Schmidt TA. Safety and efficacy of recombinant activated factor VII: a randomized placebo-controlled trial in the setting of bleeding after cardiac surgery. *Circulation* 2009; 120: 21–7.
17. Cattaneo M, Harris AS, Strömberg U, Mannucci PM. The effect of desmopressin on reducing blood loss in cardiac surgery – a meta-analysis of double-blind, placebo-controlled trials. *Thromb Haemost* 1995; 74: 1064–70.
18. Mannucci PM. Desmopressin (DDAVP) in the treatment of bleeding disorders: the first twenty years. *Haemophilia* 2000; 6(Suppl. 1): 60–7.
19. Ker K, Edwards P, Perel P, Shakur H, Roberts I. Effect of tranexamic acid on surgical bleeding: systematic review and cumulative meta-analysis. *BMJ* 2012; 344: e3054.
20. Székely A, Lex DJ. Antifibrinolytics. *Heart Lung Vessel* 2014; 6: 5–7.
21. Greco T, Zangrillo A, Biondi-Zoccai G, Landoni G. Meta-analysis: pitfalls and hints. *Heart Lung Vessel* 2013; 5: 219–25.
22. Scandroglio AM, Finco G, Pieri M, Ascari R, Calabrò MG, Taddeo D, Isella F, Franco A, Musu M, Landoni G, Alfieri O, Zangrillo A. Cardiac surgery in 260 octogenarians: a case series. *BMC Anesthesiol* 2015; 26: 15.
23. Ranucci M, Carboni G, Cotza M, Bianchi P, Di Dedda U, Aloisio T, Surgical and Clinical Outcome Research (SCORE) Group. Hemodilution on cardiopulmonary bypass as a determinant of early postoperative hyperlactatemia. *PLoS One* 2015; 10: e0126939.

### Supporting Information

Additional Supporting Information may be found in the online version of this article at the publisher's web-site:

#### Appendix S1. Ad interim analyses results.

**Figure S1.** Point estimated and 99% confidence interval for the absolute risk reduction.

**Figure S2.** Point estimated and 99% confidence interval for the relative risk.

**Table S1.** Bleeding and transfusion in the subgroup of patients randomized in theater.

**Table S2.** Bleeding and transfusion in the subgroup of patients randomized in the ICU.

# Author Query Form

Journal: AAS  
Article: 12740

Dear Author,

During the copy-editing of your paper, the following queries arose. Please respond to these by marking up your proofs with the necessary changes/additions. Please write your answers on the query sheet if there is insufficient space on the page proofs. Please write clearly and follow the conventions shown on the attached corrections sheet. If returning the proof by fax do not write too close to the paper's edge. Please remember that illegible mark-ups may delay publication.

Many thanks for your assistance.

Query reference	Query	Remarks
1	<b>AUTHOR: Please check that authors and their affiliations are correct.</b>	
2	<b>AUTHOR: Please confirm that given names (red) and surnames/family names (green) have been identified correctly.</b>	
3	<b>AUTHOR: Please check that the corresponding author address details are correct.</b>	
4	<b>AUTHOR: Please check the hierarchy of heading levels.</b>	
5	<b>AUTHOR: Please give address information for SAS Institute: town, state (if applicable), and country.</b>	