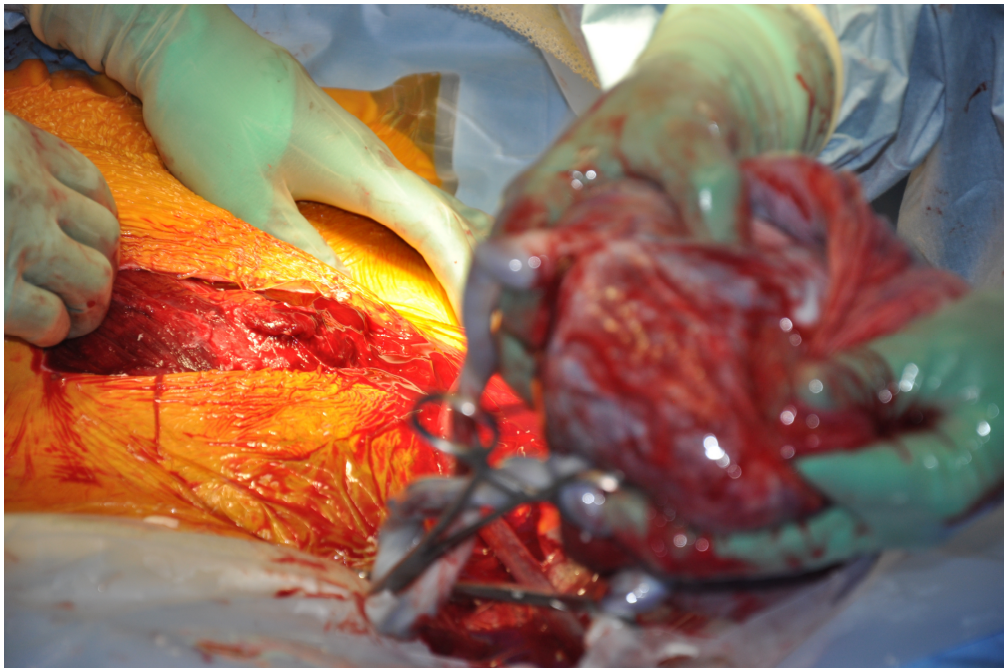
UNIVERSITY OF COPENHAGEN
FACULTY OF HEALTH AND MEDICAL SCIENCES

Aetiology and treatment of severe postpartum haemorrhage

PhD thesis

Hellen McKinnon Edwards



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This thesis has been submitted to the Graduate School of Health and Medical Sciences, University of Copenhagen September 2016

REGION Herlev og Gentofte Hospital

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SUBMITTED

December 22nd 2016

THESIS DEFENCE

March 15th 2017

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Contents

Preface

This research was carried out between June 2012 and August 2013, where I was employed as a research fellow at the Department of Anaesthesiology and Intensive Care Medicine, Herlev Hospital, University of Copenhagen, Denmark; and between May 2014 and August 2016 where I was employed as a research fellow at the Department of Obstetrics and Gynaecology, Herlev Hospital, University of Copenhagen, Denmark. I was enrolled as a PhD-student at the Graduate School of Health and Medical Sciences, University of Copenhagen.

This PhD including the Randomised Clinical Trial and research stay abroad were financially supported by:

- The Department of Gynaecology and Obstetrics, Herlev Hospital, University of Copenhagen, Denmark.
- Herlev Hospital Research Council.
- The Department of Anaesthesiology and Intensive Care Medicine, Herlev Hospital, University of Copenhagen, Denmark.
- The A.P. Møller Foundation for the Advancement of Medical Science.
- The Classenske Fideicommis Jubilee Fund.
- Travel grant from Copenhagen University
- "Kontorchef Gerhard Brøndsteds" travel grant
- Travel Grant from "Danske Lægers Forsikring under Codan/SEB pension".
- "Handelsgartner Ove William Buhl Olesen og ægtefælle fru Edith Buhl Olesens Mindelegat".
- "Gangstedfonden".
- "Hans og Nora Buchards Fond".
- "Oda og Hans Svenningsens Fond".
- "Aase og Ejnar Danielsens Fond".
- The Danish Council for Independent Research, Medical Sciences.
- The Danish Capital Region's Medical Research Foundation.
- Laerdals Acute Foundation.
- "Fonden af 17-12-1981".

None of the funding sources influenced the study designs, data collection, data management, data analyses or reporting.

Acknowledgements

Thanks to the Head of the Department of Gynaecology and Obstetrics, Helle Ejdrup Bredkjær for believing in the project and supporting me financially between funding.

Of my supervisors I firstly wish to thank Anne Wikkelsø for entrusting me with the FIB-PPH trial, for encouraging me to continue research in the field of obstetrics and anaesthesia, and for support throughout. Thanks to Jens Langhoff-Roos for believing in me, for lengthy discussions of not only my research but in obstetrics in general, for making me strive higher, and for not giving up on me. Thanks to Jens Svare for taking me on as a PhD student, for making sure rules were followed, and for always taking the time to read through my drafts even for the umpteenth time. Thanks to Jeannet Lauenborg for taking action when funds were low, for caring when times were tough, and for having an eye on detail in my drafts. Last but not least thanks to Michael Paidas for arranging my stay at Yale, and Chicago, and for continuous collaboration.

Thanks to Ann Møller, Jakob Stensballe, Arash Afshari and all collaborators in the FIB-PPH trial especially Kim Ekelund, Charlotte Albrechtsen, Lars Møller Pedersen, Mette Gøttge Madsen, and Lone Fuhrman for making me feel welcome in the project and for being on top of things throughout the trial. Furthermore thanks to Ulla, Belinda and Helle for their practical help in the trial, and for support whenever needed. To Steen Rasmussen and Tobias W Klausen for help with data even at very short notice.

I owe a huge thanks to Lars Nannestad Jørgensen for introducing me to the world of research. I am also grateful for the opportunity to have been part of two different research units as a PhD student, in both the Anaesthesia research unit and in the OBGYN research unit at Runddyssen 13, for great company, good support, and reflections on research. Especially thanks to Suzanne, Malin, Agnieszka, and Margrethe for encouragement, words of wisdom, and statistical debates.

Finally I wish to thank my fiends and family, especially Ilda, Sarah, Nina, Martin and Stefan for helping my through when times were tough, and to Veronica, Jessica, Sara and Lisa for encouragement from afar. A special thanks to Johnni's family for working on the house even when I wasn't around, and to Johnni for making us a home, and always ready to comfort when I felt most troubled.

Scientific papers included in this PhD thesis

1. Wikkelsø AJ, Edwards HM, Afshari A, Stensballe J, Langhoff-Roos J, Albrechtsen C, Ekelund K, Hanke G, Secher EL, Sharif HF, Pedersen LM, Troelstrup A, Lauenborg J, Mitchell AU, Fuhrmann L, Svare J, Madsen MG, Bødker B, Møller AM, FIB-PPH trial group. **Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized controlled trial**. BJA 2015 Apr;114(4):623-33[1].

2. Edwards HM, Wikkelsø AJ, Langhoff-Roos J, Afshari A, Møller AM, Lauenborg J, Svare JA, Stensballe J. **Massive postpartum transfusion: a multidisciplinary observational study** (awaiting new submission)[2].

3. Edwards HM, Svare JA, Wikkelsø AJ, Lauenborg J, Langhoff-Roos J. **Causes and predictors of postpartum blood loss: a cohort study** (awaiting new submission)**.**

Abbreviations

PPH	Postpartum haemorrhage
AIP	Abnormal invasive placenta
RBC	Red Blood Cell
FFP	Fresh Frozen Plasma
PLT	Platelets
TRALI	Transfusion related acute lung injury
RCT	Randomised controlled trial
CI	Confidence interval
SD	Standard deviation
IQR	Interquartile range
OR	Odds ratio
ITT	Intention-to-treat
INR	International normalized ratio
APTT	Activated partial thromboplastin time

Summary

This thesis is comprised of three studies focusing on severe postpartum haemorrhage (PPH). PPH is a major cause of maternal morbidity and mortality worldwide. Risk factors include retained placenta, prolonged duration of the third stage of labour, previous caesarean section, and operative vaginal delivery. Occurrence and development of PPH are, however, unpredictable and can sometimes give rise to massive haemorrhage or even hysterectomy and maternal death. Severe haemorrhage can lead to coagulopathy causing further haemorrhage and requiring substitution with blood transfusions.

The aim of this thesis was to investigate causes of severe PPH and investigate methods of early prevention.

The first study was a randomised controlled double-blinded trial investigating the effect of treatment with pre-emptive fibrinogen on women with severe PPH. The primary outcome was the need for red blood cell transfusion at 6 weeks postpartum. A total of 249 women were randomised to either 2 grams of fibrinogen or placebo. The mean concentration of fibrinogen increased significantly in the intervention group compared to the placebo group (0.40 g/L, confidence interval: 0.15-0.65), but there was no difference in the need for postpartum blood transfusions (relative risk 0.95, confidence interval: 0.15-1.54). No thromboembolic complications were detected.

The second study was a population-based observational study including 245 women receiving ≥10 RBCs due to PPH. The cohort was identified by combing data from The Danish Transfusion Database with The Danish Medical Birth Registry, with further data extraction and validation through review of patient charts. The main causes of massive postpartum transfusion were atony (38%) and abnormal invasive placenta (25%). Two of the women in the cohort died, an additional six had a cardiac arrest, and a total of 128 women (52%) required a hysterectomy. Hysterectomy was associated with increased blood loss, increased number of blood transfusions, a higher fresh frozen plasma to red blood cell ratio (p=0.010), and an increased number of red blood cells before first platelet transfusion (p=0.023). Hysterectomy led to haemostasis in only 70% of cases.

The third study was a register-based cohort study, including 43,357 vaginal deliveries from two large Danish maternity units. Different cut-offs were used to define PPH. There was a difference in distribution of causes depending on the cut-off used, with atony playing a decreasing role and a retained placenta an increasing role the higher the cut-off used. In a multivariate linear regression model retained placenta was identified as a strong predictor of quantity of blood loss. The duration of the third stage of labour was a very weak predictor after adjusting for the influence of a retained placenta.

In conclusion, an improved diagnosis of the causes of PPH especially retained placenta, together with an early recognition and treatment of coagulopathy, seem to be important in reducing severe PPH in an aim to minimize associated maternal morbidity.

Summary in Danish (Resumé)

Denne afhandling udgøres af tre studier, der fokuserer på svær postpartum blødning (PPH). PPH er én af de væsentligste årsager til maternel morbiditet og mortalitet i hele verden. Risikofaktorer inkluderer retineret placenta, forlænget varighed af fødslens tredje stadie, sectio antea og instrumental forløsning. Forekomst og udvikling af PPH er dog uforudsigelig, og kan af og til medføre massiv blødning, eller endda hysterektomi og maternel død. Svær blødning kan føre til koagulopati med yderligere blødning til følge og behov for substitution med blodtransfusioner.

Formålet med denne afhandling var at undersøge årsagerne til svær PPH, samt undersøge metoder til tidlig forebyggelse.

Det første studie var et randomiseret kontrolleret dobbelt-blindet studie, der undersøgte effekten af tidlig behandling med fibrinogen til kvinder med svær PPH. Det primære outcome var behov for SAGM transfusioner op til 6 uger postpartum. Vi randomiserede 249 kvinder til enten 2 g fibrinogen eller placebo. Koncentrationen af fibrinogen steg signifikant i interventions gruppen i forhold til placebo gruppen (0.40 g/L, konfidens interval: 0.15-0.65), men der var ingen forskel i behovet for postpartum blodtransfusioner (relativ risiko 0.95, konfidens interval: 0.15-1.54). Der blev ikke observeret tromboemboliske komplikationer.

Det andet studie var et populations-baseret observationelt studie med 245 kvinder, der modtog ≥10 portioner erythrocytkoncentrat (SAGM) pga. PPH. Kohorten blev identificeret, ved at kombinere data fra Dansk Transfusionsdatabase med Fødselsregisteret, med yderligere data ekstraktion og validering ved struktureret journal gennemgang. De hyppigste årsager til massiv postpartum transfusion var atoni (38%) og abnormt invasiv placenta (25%). I kohorten, var der to kvinder, der døde, yderligere seks fik hjertestop, og 128 kvinder (52%) havde behov for hysterektomi. Hysterektomi var associeret med øget blodtab, øget antal blod transfusioner, en højere ratio af frisk frosset plasma til SAGM (p=0.010), og et øget antal SAGM før første trombocyttransfusion (p=0.023). Hysterektomi førte kun til hæmostase i 70% af tilfældene.

Det tredje studie var et register-baseret kohorte studie, der inkluderede 43.357 vaginale fødsler fra to store danske fødeafdelinger. Der blev brugt forskellige cut-offs til at definere PPH. Der var en

forskel i distributionen af årsager afhængig af hvilken cut-off, der blev brugt, hvor atoni spillede en aftagende rolle og retineret placenta en stigende rolle jo højere cut-off, der blev sat. I den multivariate liniære regressions model blev retineret placenta identificeret som en stærk prædiktor for mængden af blodtab. Efter justering for indflydelsen af en retineret placenta var varigheden af fødslens tredje stadie en svag prædiktor.

En forbedret diagnosticering af årsagerne til PPH, især retineret placenta, og en tidlig erkendelse og behandling af koagulopati, er vigtige for at reducere de maternelle komplikationer ved svær PPH.

Introduction

Postpartum haemorrhage (PPH) plays a significant role in maternal morbidity and mortality, and has had an impact on the world for centuries[4,5]. Thousands of women die each year due to PPH, a few of which have given rise to not only cultural and medical innovations, but also shaped the history of the world. The Taj Mahal was built by Mughal emperor Shah Jahan in memory of his wife that died of PPH in 1631after giving birth to their 14th child[6]. Princess Charlotte, daughter of King George IV of England, was in 1817 the only eligible heir to the throne, but died after stillbirth due to 50 hours of labour and PPH, leading to change of reign and the birth of the future Queen Victoria[7]. Last but not least in 1825 the British obstetrician James Blundell was the first to successfully transfuse human blood. He saved the life of a woman with PPH, by using blood from the woman's husband; later he went on to invent several instruments for transfusion[8–10].

Sadly these innovations do not nearly weigh up the tragedy of a maternal death, and even though maternal deaths worldwide are decreasing[11], PPH has shown an increasing trend over the last few years to an incidence of 3-8% in the developed world[12–14], and is the most common cause of maternal morbidity[15,16]. Therefore, research in prevention and treatment of PPH including recovery measures for women developing life-threatening haemorrhage is needed more than ever[17].

Background

Postpartum haemorrhage – aetiology and risk factors

PPH is traditionally defined as blood loss ≥500 ml in the first 24 hours following childbirth, often developing minutes after childbirth, but can also be secondary if occurring after the first 24 hours up to 6 weeks postpartum[18]. For women undergoing caesarean section the cut-off is higher and usually defined as ≥1,000 ml[19]. However, not all countries or studies agree on these definitions, creating not only confusion but also conflicting results[20]. Further inconsistency is found when it comes to defining severe PPH, where there is variation in not only the cut-off used to define it, but also no uniform agreement of whether to use the term severe, major or moderate PPH[20–23]. Estimation of blood loss can be assessed in many ways depending on the equipment available. Visual estimation is the easiest method, but also the method that is most inaccurate as large quantities of blood loss are often underestimated and small quantities of blood loss overestimated compared to blood collection bags or weighing of drapes ect[4,18,24].

The aetiologies of PPH are classically divided into four different categories, known as the four T's – Tone, Trauma, Tissue, and Thrombin[21]. Tone refers to atony, which is insufficient contraction of the uterus during and after delivery of the placenta, leading to extensive bleeding from the placental bed. Trauma refers mainly to lacerations of the vagina and perineum, graded from first to fourth degree depending on their depth and extent, but can also include vulvar and vaginal haematomas or uterine rupture, all of which will need surgical repair. Tissue refers to retained placenta or fragments of placenta inhibiting contraction of the uterus. Thrombin refers to coagulopathies, that can be defects known prior to childbirth or developed during or after childbirth due to other complications such as amniotic fluid embolism[19,21]. The majority of cases are traditionally attributed to atony[21].

The time from the delivery of the baby to the delivery of the placenta is known as the third stage of labour[25]. The uterus will under normal circumstances contract and expel the placenta within 10 minutes[26–28], efficiently cutting off the blood flow to the placenta[18]. The placenta will in some circumstances need manual removal if it is not delivered spontaneously. If the duration of the third stage of labour exceeds 30 minutes there is an increased risk of PPH[25,29,30]. Recent studies have questioned the 30 minute threshold and have suggested that the risk of PPH is increased

after only 15 to 20 minutes[26–28,31,32]. An active management of the third stage of labour has been shown to reduce the risk of PPH. This includes administration of oxytocin (a uterotonic that stimulates contraction of the uterus), controlled cord traction and uterine massage[33]. If the placenta is not delivered spontaneously, several conditions should be considered. The placenta could be detached from the uterine wall but still trapped inside the uterus due to a closed cervix: *an entrapped placenta*; the placenta is not detached but there are no signs of invasive growth in the uterine wall: *an adherent placenta*; or there is abnormal invasive growth into or through the uterine wall: *an abnormal invasive placenta (AIP)*[34,35].

AIP has an incidence of approximately 0.2-3 per 1,000 deliveries[36–39]. Depending on the depth of attachment AIP is termed: *placenta accreta* (placenta attached to the myometrium); *placenta increta* (placenta invades the myometrium); or *placenta percreta* (placenta invades through the myometrium)[36,40]. AIP often leads to severe PPH requiring blood transfusions and in more severe cases even the need for hysterectomy, complications that can be minimized if diagnosed before labour[39]. Currently, up to 50% of AIP cases are identified antenatally through ultrasound screening of women with a prior caesarean section and placenta praevia[36,41].

Numerous epidemiological studies have been performed to try and identify women at risk of developing PPH, in the hope of initiating sufficient preventive measures[30,42–44]. Some of the risk factors identified include multiparity, previous caesarean section, hypertensive disorders, macrosomia, previous PPH, induction of labour, augmentation of labour, operative vaginal delivery, caesarean section and placenta praevia[45,46]. Some of the risk factors have a higher risk of PPH than others, but women with high risk or multiple low risks can still have a completely uncomplicated delivery[43]. Furthermore, 22-39% of women that develop PPH have no risk factors, making it extremely difficult to predict which women will in fact develop PPH[12,47–49].

There are a wide range of complications following PPH. Mild cases of PPH can lead to anaemia, fatigue, depression and feelings of separation or anxiety[21,50,51]. In more severe cases the complications are often critical and involve blood transfusions, open surgery, organ failure, treatment in an intensive care unit, thromboembolic complications, hysterectomy and in worst case even death[12,52–54].

Haemostasis in pregnancy and postpartum

Haemostasis is the process that maintains equilibrium between coagulation and fluidity of blood in damaged blood vessels through the actions of the coagulation cascade, platelets, and fibrinolysis[55,56]. The purpose of the coagulation cascade is to stop bleeding by forming a clot, through a cascade of processes initiated after the exposure of tissue factor primarily after vascular damage[57,58]. The coagulation system is comprised of clotting factors in an inactive state that become activated through a cascade of processes, and culminates with conversion of large amounts of thrombin from prothrombin. Thrombin converts fibrinogen into fibrin fibres, which together with activated platelets and von Willebrand factor create the blood clot[56,59]. Fibrinogen is a glycoprotein synthesized in the liver and is indispensable in formation of the clot not only through conversion to fibrin fibres but also for platelet aggregation.[60] There are several regulators of the coagulation cascade including the anticoagulation factors: antithrombin, protein C, and protein S that limit the formation of clots in healthy vessels[56,58]. In addition simultaneous activation of fibrinolysis dissolves the clot in a highly regulated process, preventing excessive clot formation[58,59].

During pregnancy blood volume and coagulation increase while anticoagulants and fibrinolysis decrease, all part of the prophylactic measures to prepare for blood loss and placental separation after childbirth[55,58]. This change in haemostasis involves a rise in some of the coagulation factors including prothrombin, fibrinogen, and von Willebrand factor, but also a decrease in platelet count due to haemodilution and presumed consumption at the placental site[55,61,62]. However, this hypercoagulable state in pregnancy leads to an up to six-fold increase in the risk of thromboembolic complications including pulmonary embolisms and deep vein thrombosis[62,63]. Additional increase in coagulation factors including fibrinogen takes place during labour and delivery. Coagulation factors are activated through release of abundant amounts of tissue factor upon placental separation leading to formation of clots. Increased levels of fibrinogen and platelets postpartum are also a result of inflammation[55]. An unimpaired coagulation system will together with sufficient contraction of the uterus result in minimal blood loss after delivery[55,61,62]. However, a high consumption of coagulation factors and platelets in formation of clots at the placental site can potentially lead to depletion if haemorrhage is ongoing[62,64]. Under normal circumstances coagulation factors remain high the first few days after delivery with fibrinolysis rising to normal levels within 1-2 days postpartum and normal coagulation attained within 4-6 weeks postpartum.[55,61]

Massive haemorrhage and transfusion

Massive haemorrhage is defined as loss of total blood volume within 24 hours, 50% within 3 hours, or a rate of blood loss of 150 ml/min[65]. Blood loss of this quantity can be difficult to assess during an emergency situation, which is why massive haemorrhage can also be defined as haemorrhage requiring massive transfusion of ≥10 units of red blood cells (RBC) within 24 hours[66]. In obstetrics there is no well-defined consensus for massive haemorrhage, with the terms major and massive PPH being used at random for blood loss of more than 1,000 ml to blood loss of more than 2500 ml[21,67–69].

Massive haemorrhage following trauma, surgery or childbirth may lead to coagulopathy – a state of impaired haemostasis. In all three circumstances the abundant release of tissue factor leads to activation of the coagulation cascade and consequent consumption of coagulation factors and platelets[57,70]. The simultaneous systemic hypoperfusion causes hypothermia and acidosis, inhibiting coagulation and activating anticoagulation factors and fibrinolysis, which complicate coagulation further[66,70]. At the same time transfusion with RBCs, crystalloids or colloids are given in an effort to re-establish perfusion causing additional dilutional coagulopathy. The combination of consumptive and dilutional coagulopathy, acidosis and hypothermia, known as the lethal triad will result in further haemorrhage[57,70,71]. Therefore, treatment involving not only volume resuscitation and surgical control of haemorrhage, but also correction of coagulopathy is necessary[71,72].

Fibrinogen is the first coagulation factor known to drop to critical levels during massive haemorrhage, and as the normal level of fibrinogen is 2.0-4.5 g/L in healthy adults, low levels are difficult to substitute with FFP alone, where the concentration is 1-3 g/L[60,73]. Additional substitution is, however, possible through cryoprecipitate and fibrinogen concentrate. Cryoprecipitate contains high concentrations of fibrinogen (approximately 15 g/L), von Willebrand factor and other coagulation factors. However, cross matching and thawing is necessary before administration. Fibrinogen concentrate on the other hand only contains fibrinogen (15-20 g/L), and comes as a powder that only requires dissolving in sterile water before administration[60,74].

Identifying patients with low levels of specific coagulation factors is possible through conventional laboratory testing. However, these tests can be time consuming and they do not assess the general functionality of coagulation, which is why point-of-care viscoelastic assays are being used more and more. These assays can be performed bedside and give an assessment of clot formation and fibrinolysis, thereby providing vital information on the development of coagulopathy[66,75]. Prevention and treatment of coagulopathy in patients with massive haemorrhage is also possible with early transfusion of RBCs, FFP and PLTs. Furthermore, studies from both trauma and non-trauma have shown a reduction in mortality when a fixed ratio of 1:1:1 of PLTs, FFP and RBCs is used during massive haemorrhage[76–78].

Due to the highs risks associated with blood transfusions, all strategies that can reduce blood transfusions are essential. Today the risk of transmission of infection through blood transfusions is low; instead the risks are related to non-infectious reactions including haemolytic, allergic, and immunological reactions that occur in approximately 1% of all transfusions[79–81]. Transfusion related acute lung injury (TRALI) is an immunological reaction and the leading cause of transfusion related morbidity and mortality with an incidence of 0.08-15%[79]. TRALI evolves within 6 hours of transfusion and is mainly associated with plasma transfusions. Symptoms include dyspnoea, hypoxaemia and hypotension due to pulmonary oedema and up to 70% will need respiratory support[79]. Additional complications are seen in patients requiring massive transfusions, including metabolic complications due to haemolysis and high levels of citrate, and transfusion associated circulatory overload[66].

Severe postpartum haemorrhage – prevention and treatment

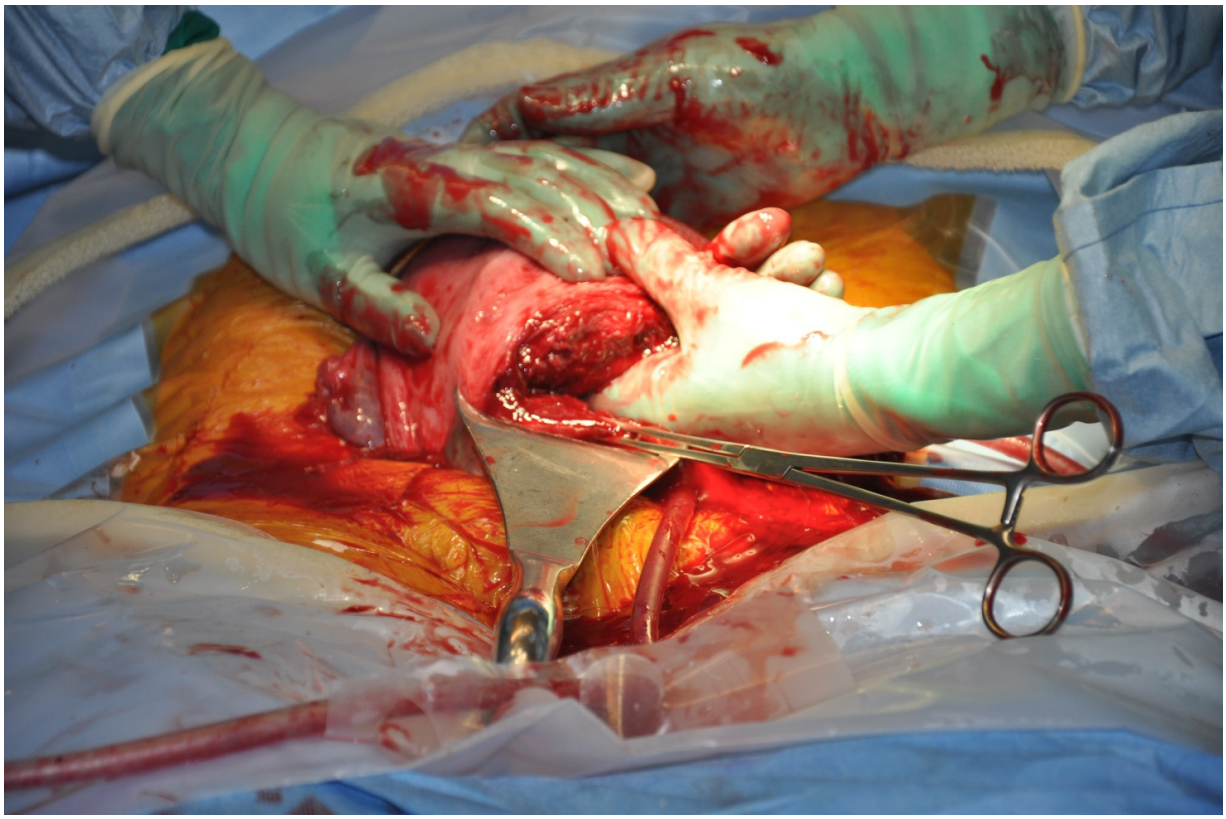
Active management of the third stage of labour and removal of a retained placenta can reduce the risk of PPH. Further preventive measures include minimizing avoidable risk factors or giving additional uterotonics to high risk women[17,21,82]. Once PPH has developed treatment options relate to the cause of haemorrhage: uterotonics for atony, surgical repair of lacerations, removal of retained tissue, and correction of diagnosed coagulopathy[21]. However, progression in severity is not always avoidable, and has therefore led to increased focus on early warning signs and treatment of severe PPH. Risk factors associated with progression to a more severe PPH include instrumental delivery, augmentation of labour, multiple pregnancy, polyhydramnios and hypertensive disorders[42,83]. As these risk factors are not always preventable or directly treatable, recent studies have tried to identify more specific predictors of severity related to coagulopathy.

The main focus has been on fibrinogen since Charbit et al in 2007 showed that a fibrinogen concentration ≤2 g/L was 100% predictive of severe PPH[84]. The study included 128 women with PPH of which 50 (39%) developed severe PPH (defined as haemoglobin decrease ≥4 g/dl, transfusion of ≥4 RBCs, embolization, arterial ligation, hysterectomy or death). Women were enrolled if they had PPH requiring IV prostaglandin infusion (uterotonics). A fibrinogen level of ≤2 g/L at enrolment was identified in 11 of the 50 women (22%) that developed severe PPH. A number of other studies have confirmed the association between low levels of fibrinogen and blood loss in PPH[23,85,86]. However, association is not always the same as causation. The results from Charbit et al have therefor led to recent studies investigating the impact of fibrinogen substitution on development of a more severe PPH[87–89]. However as the normal level of fibrinogen at delivery is higher than in the non-pregnant woman (3.5-6.5 g/L vs. 2.0-4.5g/L), the exact threshold for intervention is unclear[61,73,90].

Intensive treatment and care becomes the main focus once PPH has progressed, involving a close collaboration between obstetricians, gynaecologists, anaesthetists and sometimes also coagulation experts. Atony is mainly treated with additional uterotonics, but other causes of PPH should be considered if haemorrhage is refractory to first-line uterotonics[33,91]. Further treatment of all causes of ongoing PPH mainly takes place in the operating room involving all of the multidisciplinary team. Surgical repair of lacerations, removal of placental tissue and intrauterine balloon tamponade can be performed from a vaginal approach. Additional surgical interventions require laparotomy, with uterine haemostatic suturing (e.g. B-lynch suture) or artery ligation being attempted before hysterectomy[21,91,92]. Even though hysterectomy is often considered last option in uncontrollable PPH, it does not necessarily lead to haemostasis perhaps due to untreated coagulopathy[54,93,94].

Coagulopathy should be considered early on in the events of progressing PPH, with simultaneous focus on both transfusions and surgical control as neither can stand alone[4,95]. As the rate of transfusion in obstetrics is relatively low at 0.5-2.0%, research into the optimal ratio of RBCs, FFP and PLTs is scarce[44,96–98]. A few retrospective studies have shown that a high FFP:RBC ratio was associated with a reduced risk of interventions and a higher success rate of hysterectomy, but none of the studies were in relation to PPH requiring massive transfusion[94,99]. The methods used to monitor coagulopathy in severe PPH are the same as in other patients with severe haemorrhage. The Danish guideline for PPH recommends traditional laboratory tests

including platelet count, international normalized ratio (INR), activated partial thromboplastin time (APTT) and fibrinogen early on in the course of events, or if available point-of-care viscoelastic assays[82]. Laboratory tests do not give rapid results, and the haemostasis of the patient can have changed substantially before it is possible to react to the results[90]. It is therefore of great importance to be continuously aware of formation of clots in the operating field.



Objectives

Through the studies included in this PhD thesis we aim to investigate the causes of severe postpartum haemorrhage and minimize the proportion of women developing severe postpartum haemorrhage by identifying methods for early prevention.

The objectives and hypotheses of this thesis were:

- To assess if pre-emptive treatment with fibrinogen concentrate could reduce the need for red blood cell transfusion in relation to postpartum haemorrhage *(Study I)*.
    - The hypothesis was that pre-emptive treatment with fibrinogen could reduce the need for red blood cell transfusion in women with postpartum haemorrhage.

- To describe the severe cases of PPH requiring massive transfusion including the surgical procedures leading to bleeding control, the causes, and the complications associated with the need of massive postpartum transfusion. And to describe the subgroup of women treated with hysterectomy and compare the use of RBC, FFP, and PLT transfusions with women not treated with hysterectomy during massive postpartum transfusion *(Study II)*.
    - The hypothesis was that women treated with a hysterectomy due to massive postpartum transfusion had received less or late fresh frozen plasma and platelets compared to women with massive postpartum transfusions not treated with a hysterectomy.

- To investigate whether the distribution of causes of postpartum blood loss depended on the cut-off used to define PPH. And to investigate the association between quantity of postpartum blood loss, the duration of the third stage of labour, a retained placenta, and other risk factors *(Study III)*.
    - The hypothesis was that the distribution of causes of postpartum blood loss depended on the cut-off used to define PPH and that a retained placenta had a higher impact on the quantity of blood loss than the duration of the third stage of labour.

Material and methods

Choosing the correct study design depends not only on the research question, but also on practical issues including the setting, how common the condition is, the time frame available, data available, and funding[100]. A randomised control trial (RCT) can lead to high quality evidence of treatment effect, and was the method of choice for study I[1]. Observational studies have a higher risk of confounding but can be used to identify potential associations between exposures and outcome[101]. We chose to address the research questions in study II and III through observational studies, identifying two separate cohorts from different registries. Thereby, taking advantage of some of the exclusive data already available in Danish registries, where data on more or less all residents is registered, due to all being assigned a unique Civil Registration Number [102,103].

National registries

The Danish National Patient Registry

The Danish National Patient Registry was established in 1977 and consists of data from all hospitalised patients[104]. It is compulsory for all hospitals in Denmark to report to the registry, and the registry is the general core database for health issues in Denmark and is a source for some of the other more specialised registries and databases such as The Danish National Birth Registry and The Danish Transfusion Database[104].

The Danish National Birth Registry

The Danish National Birth Registry contains information regarding all births in Denmark dating back to 1973. The registry has since 1995 received all information regarding maternal demographics, parity, pregnancy, labour and delivery by combining the unique Civil Registration Numbers of both mother and child with data from The Danish National Patient Registry[105].

The Danish Transfusion Database

The Danish Transfusion Database receives information directly from regional blood banks and The National Patient Registry, and has done so since 1997. However, full coverage of Denmark was not complete until 2005. The database includes information regarding allogenic transfusions

including data on the recipient, serial numbers of the blood products and time of delivery of blood products[106].

The Copenhagen Obstetric Database

The Copenhagen Obstetric Database was established in 1996 and receives detailed information on maternal demographics, pregnancy, labour, delivery and details on the new-born directly from midwives and specialist doctors during and after discharge. It has a very high internal validity, and includes additional information that is not registered in The Danish National Patient Registry such as the quantity of blood loss[107].

Study populations

All of our studies were comprised of women with an assorted range of severity of PPH.
Study I was a randomised controlled double-blinded study where the primary outcome was the need to transfuse RBCs up to six weeks postpartum in women randomised to either placebo or 2 grams of fibrinogen concentrate. A dose of 2 g was chosen based on an average weight of 65.9 kg, with a target fibrinogen level of 4 g/L from a mean fibrinogen level of 3.4 g/L after 500-1,000 ml of postpartum blood loss[87]. Fibrinogen concentrate was given at time of inclusion, and without taking body weight or fibrinogen levels (pre-emptive) into account, to ensure quick administration and in accordance with our objectives. Secondary outcomes included total blood loss, total number of RBCs transfused, haemoglobin <58 g/L, and severe PPH (defined as decrease in haemoglobin >40 g/L, transfusion of ≥4 RBCs, embolization, arterial ligation, hysterectomy or death). Furthermore, an important part of the trial was monitoring of haemostasis and adverse events related to fibrinogen concentrate. Inclusion criteria were women with PPH ≥500 ml requiring manual removal of placenta after vaginal delivery or PPH ≥1,000 ml after caesarean section or requiring exploration of the uterus after vaginal delivery within 24 hours of delivery. Exclusion criteria were: known inherited coagulation deficiencies, antenatal anti-thrombotic treatment, pre-pregnancy weight <45 kg, or refusal to receive blood transfusions. A multicentre approach at four university affiliated hospitals in the Capital Region of Denmark was decided on, due to the relatively low incidence of PPH and the plan to include 245 women over a two year period[1,87]. The trial was designed as a superiority trial and the sample size was based on the fact that approximately 1% of women giving birth receive blood transfusions and 1.75% have blood loos > 1,000 ml, thereby the incidence of transfusion in PPH > 1,000 ml is 57%[108,109]. With an estimated risk reduction of 33%, α=0.005 and 80% power, we would need to include 107 women in each group. This would lead to a requirement of 245 women, if

calculating with a 15 % dropout/missing data. A follow-up period of 6 weeks was chosen to monitor re-bleeding/secondary PPH that is defined up to 6 weeks postpartum; and to monitor thromboembolic complications, where there is a known increase in risk up to 6 weeks postpartum[63].

Study II was an observational study where we investigated the influence of transfusions on hysterectomy in women with massive postpartum transfusion. To be able to gain a sufficient cohort size, we included women from all over Denmark for a 9-year period from 2001 to 2009. Women were identified by combing data from The Danish National Birth Registry and The Danish Transfusion Database, and included if they received ≥10 units of RBCs within a 24 hour period up to 6 weeks postpartum. In order to gain sufficient information regarding transfusions, causes, and procedures it was necessary to review all patient charts and extract relevant data. Deliveries from hospitals not included in The Danish Transfusion Database before 2005 were excluded together with women without accessible patient charts, and women receiving blood transfusions due to non-obstetric causes[2].

For our final study, study III, the primary outcome was quantity of postpartum blood loss, where we investigated the distribution of causes and the effect of a retained placenta and the third stage of labour. We used data from The Copenhagen Obstetric Database and included all vaginal deliveries from 22 to 43 weeks of gestation from 2009 to 2013, in order to obtain a large cohort with a high degree of variation in quantity of blood loss. We excluded all cases with blood loss below 50 ml due to interpretation of faulty registration, and all hospitals reporting to the registry for less than one year[3]. We calculated the duration of the third stage of labour from the time of delivery of the neonate until the time of either spontaneous delivery of the placenta or manual removal of the placenta. A retained placenta was defined as diagnosis of AIP, retained placenta or manual removal of either placenta or tissue. The diagnosis “retained placenta” is not used if the placenta is delivered spontaneously.

When comparing causes with different definitions of PPH (Study III), and between the three studies, each patient was only assigned a single cause. “Retained placenta/tissue” was given as the primary cause for women with retained placenta, retained tissue or AIP. “Lacerations” was given as the primary cause for women without “retained placenta/tissue” and with lacerations of the cervix, vagina or perineum including an episiotomy and paravaginal haematomas. “Other,

including atony" was given as the primary cause for women without "retained placenta/tissue" or "lacerations".

Coordinating a randomized controlled trial

The randomised multicentre double blinded clinical trial (Study I) was initiated by Dr. Anne Juul Wikkelsø[87]. I was appointed project coordinator after the first patients had been enrolled in the trial, and became responsible for all major aspects of the project for the remainder of the study period. This involved all practicalities, accountability for all patient consents, coordinating blood tests 24/7, securing additional funding, and responsibility for upholding regulations from The Department of Good Clinical Practice, The Ethics Committee, and The Danish Health and Medicines Authority. The anaesthetist was responsible for patient consent, trial drug administration and primary data collection. Other personnel groups also played a large role in the study including anaesthetic nurses taking care of randomisation, drug dispensation, and blood sampling; and obstetricians and midwives supplying information regarding the trial to as many women as possible before delivery. The project coordinators' main focus regarding training of these four different personnel groups was therefore on adherence to protocol especially regarding informed consent, randomisation, and blinding.

Informed consent

Informed consent is required by law according to the Helsinki declaration regarding participation in research studies[110]. For our study, informed consent was obtained either before delivery during preparation for a caesarean section or an epidural, or after delivery in the emergency situation when PPH requiring intervention had been determined. Obtaining informed consent during an emergency situation is known to be difficult, perhaps even more so after delivery in a situation of anxiety and pain[111–113]. The study grouped had sought approval from the local Ethics Committee regarding possibility of surrogate consent, but this had been rejected[114]. It was, therefore, crucial that I had focus on all formalities regarding the informed consent. Furthermore, all included women were asked about their experiences regarding the inclusion process in the trial during follow-up.

Randomisation

Randomisation together with allocation concealment are the most essential factors in controlling for confounders and eliminating selection bias[115]. In our study the randomisation process was

computer generated by a third party company before initiation of the study, and was stratified by centre and in blocks of four to optimise the control for confounders. Furthermore treatment allocation was concealed by using a centralised service with concealed envelopes. Once randomised, personnel not involved in the treatment of the patient dispensed placebo or fibrinogen concentrate in opaque syringes, thereby concealing allocation to all personnel in charge of further treatment[87].

Blinding

Blinding is used to control for information bias, where interpretation of results otherwise can be influenced by knowledge of allocation[116]. Triple-blinding was a fundamental part of the study's protocol and involved blinding of patients, personnel involved in treatment, trial investigators, and statisticians[87]. Blinding of fibrinogen measurements was also necessary to prevent clinicians from identifying patients with increasing levels of fibrinogen after infusion of the study drug. Therefore, all fibrinogen analyses at inclusion were analysed by a separate laboratory. To assess the success of blinding we asked all primary anaesthetist involved in the inclusion process and all included women whether they had suspicion of treatment allocation and why.

Fibrinogen measurements

All blood samples for fibrinogen were collected, frozen and stored during the study period. After completion of the study all samples were analysed using the Clauss method[117] at one single laboratory, thereby eliminating methodological issues related to different testing. The normal lower limit of fibrinogen was set at 3.7 g/L[a],[118] but the threshold for hyopfibrinogenaemia was set at 2.0 g/L in accordance with the findings from Charbit et al.[84]

[a] The lower normal limit was chosen based on a review of normal reference intervals for women during pregnancy[118]. Since then a new study has estimated the normal reference interval during partus at 3.5-6.5 g/L[61]

Variables

The three studies had different data available, but the majority of variables used are the same. The variables included in each study are listed in Table 1.

Table 1. List of all variables includes in study I, II and III.

Variable	Study I	Study II	Study III
Maternal age		X[a]	X
Parity	X	X[a]	X
Gestational age at delivery	X	X[a]	X
Previous Caesarean section	X	X[a]	X
Multiple gestation	X	X[a]	X
Hypertensive disorders	X	X[a]	X
Antepartum haemorrhage	X	X	X
Previous postpartum haemorrhage	X	X[a]	
Fertility treatment		X	
Amniotic fluid abnormalities		X	X
BMI before pregnancy	X	X[a]	
Maternal weight at term	X		
Birthweight	X	X[a]	
Time and date of birth		X	X
Duration of labour			X
Induction of labour		X[b]	X
Augmentation of labour		X[b]	X
Time of birth of the placenta			X
Site of birth	X	X	X
Placenta praevia	X	X[a]	X
Abnormal invasive placenta	X	X[a]	X
Foetus presentation			X
Preterm premature or premature rupture of membranes			X
Neonatal outcome		X	
Episiotomy	X	X[b]	X
Retained tissue	X	X[c]	X
Placental abruption	X	X[b]	X
Uterine rupture	X	X[b]	X
Genital tract lacerations	X	X[c]	X
Mode of delivery	X	X[b]	X

Reason for non-vaginal delivery		X	
Epidural analgesia	X		X
Fever during labour			X
Shoulder dystocia			X
Uterine inversion	X	X	X
Quantity of blood loss	X *	X	X
Blood transfusions	X **	X	
Other causes of postpartum haemorrhage		X	
Time of start of bleeding	X	X	
Time of haemostasis		X	
Cumulated use of colloids or crystalloids	X *	X	
Cumulated use of uterotonics	X *	X	
Cumulated use of fibrinogen, tranexamic acid, or recombinant factor VIIa	X *	X	
Surgical procedures performed	X	X	
Timing of consent	X		
Blood pressure and heart rate	X *		
Measurements of fibrinogen, haemoglobin and platelets	X *		
Postoperative complications	X	X	
Adverse events after study drug	X		
Thromboembolic complications	X	X	
Death	X	X	
Assessment of blinding	X		

* Measured at inclusion and 15 minutes, 4 hours and 24 hours after study drug.
** Measured at inclusion and 15 minutes, 4 hours, 24 hours, 7 days, 6 weeks postpartum.
[a] Antenatal risk factors (age >35, BMI >35, Parity >3, birthweight >4,000g, Gestational age >42 weeks.
[b] Labour and delivery risk factors (mode of delivery = caesarean section or operative vaginal delivery)
[c] Postpartum risk factors

Statistical analyses

The results of study I were analysed as an intention-to-treat (ITT) population and as a per-protocol population. The ITT analysis was performed prior to disclosure of allocation and included all participants with informed consent that had been randomised, irrespective of whether they fulfilled exclusion criteria or whether they received the complete allocated treatment. The per-protocol analysis was performed after disclosure of allocation and excluded any participants that fulfilled exclusion criteria or received incomplete allocated treatment. Chi^2

test was used for binary outcome measures and Student's t-test or Wilcoxon rank-sum test for continuous measures of unadjusted analyses. These results are presented as relative risks with a 95% confidence interval (CI). Logistic regression analysis was used for adjusted analyses of association between baseline variables, allocated treatment, stratification variable (centre) and primary outcome. Logistic regression analysis was also used for post hoc analysis of association between significant baseline variables including stratification variable (centre) and primary outcome. These results are presented as odds ratios (OR) with a 95% CI. Changes in fibrinogen and haemoglobin levels during the first 24 hours after study drug infusion were compared between groups using longitudinal analysis (mixed-effect model).

For study II we used univariate logistic regression analyses to evaluate differences between women treated with and without hysterectomy. Analyses used to determine differences included Chi^2 test for categorical variables, t-test for normally distributed continuous variables, and Kruskal-Wallis for non-normally distributed continuous variables.

The outcome measure of quantity of blood loss in study III was logarithmic transformed (log10) due to substantial skewed non-normal distribution. Univariate and multivariate linear regression analysis were used to evaluate variables and their influence on quantity of postpartum blood loss. These results are presented as β-coefficients and 95% CI. Interpretation is quite simple: you obtain the percent change in the predicted quantity of postpartum blood loss for each variable by raising 10 to the power of the β-coefficient and subtracting 1.00.

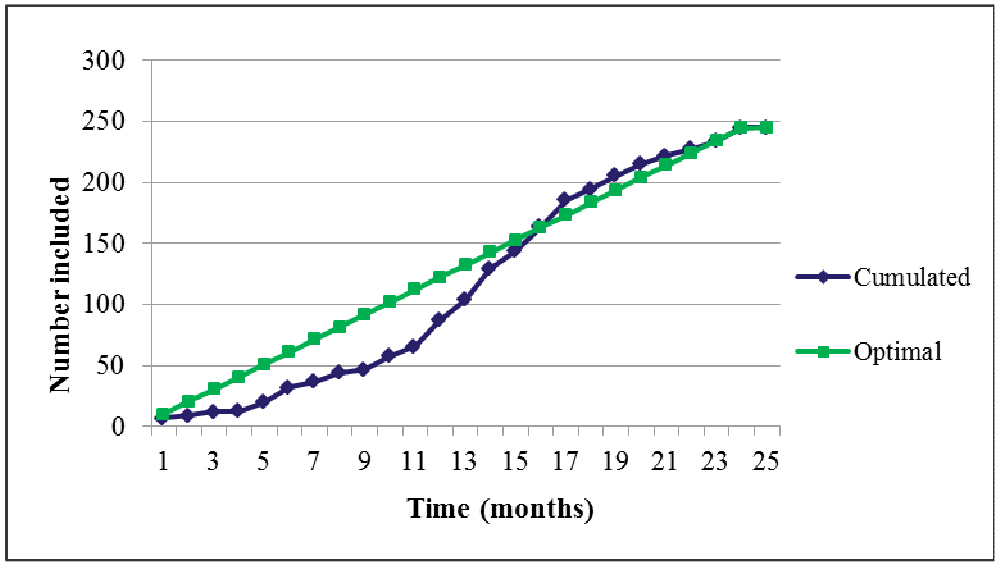
For all studies a two-sided p-value of <0.05 was considered statistically significant. All data analyses were carried out using either R statistical software (R Foundation for Statistical Computing, Vienna, Austria) or SPSS 22.0 (SPSS, Chicago, IL, USA).

Results

Study I: Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized controlled trial

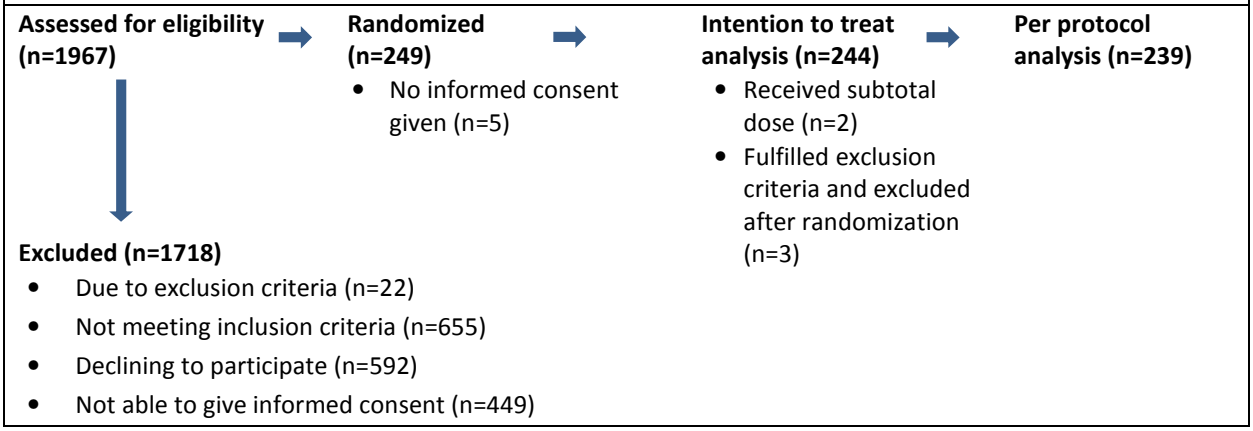
This was the first published RCT with fibrinogen concentrate in obstetrics. A total of 249 women were randomised during the planned study period of two years (Figure 1).

Figure 1. Optimal and cumulated inclusion rates of patients.



No-one was lost to follow-up but five women were excluded due to insufficient informed consent, three of whom did not receive intervention. This left 244 women for the ITT analysis; 123 in the fibrinogen group and 121 in the placebo group (Figure 2). The mean estimated blood loss at inclusion was 1,459 ml (SD ±476) with the majority (84%) included after vaginal delivery and due to retained placental tissue (64%). The mean fibrinogen concentration at inclusion was 4.5 g/L, with 2.2% below 2 g/L. The fibrinogen concentrate dose of 2 g/L corresponded with a dose of 26mg/kg and significantly increased the fibrinogen concentration 0.40 g/L (CI: 0.15-0.65) compared to the placebo group 15 minutes after administration.

Figure 2. Flow diagram of enrolment. (*Adapted from "Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized control trial"*[1]*).*



A total of 25 (20.3%) of the fibrinogen group and 26 (21.5%) of the placebo group received a RBC transfusion during the 6 week follow-up, with no significant difference in RBC transfusion at any time point registered (Table 2). The majority of women received their blood transfusions within the first 24 hours, and all first transfusions were initiated within the first week. We found no significant difference between the two groups in regard to any of the remaining secondary outcomes (Table 2).

Table 2. Unadjusted analysis of primary and secondary outcome measures. Intention to treat analysis. (*Adapted from "Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized control trial"*[1]*).*

Outcome	Fibrinogen	Placebo	Relative risk (95% CI)	p-value
Any RBC transfusion at 6 weeks	25 (20.3%)	26 (21.5%)	0.95 (0.58-1.54)	0.88*
Any RBC transfusion at 4 hours	4 (3.3%)	10 (8.3%)	0.39 (0.13-1.22)	0.11*
Any RBC transfusion at 24 hrs	14 (11.4%)	19 (15.7%)	0.72 (0.38-1.38)	0.35*
Any RBC transfusion at 7 days	25 (20.3%)	26 (21.5%)	0.95 (0.58-1.54)	0.88*
Total number of RBCs	0 [0-0]	0 [0-0]		0.83**
Post-intervention estimated blood loss	1,700 [1,500-2,000]	1,700 [1,400-200]	66 [-78;210]	0.37***
Severe PPH	20 (40.0%)	24 (52.2%)	0.77 (0.49-1.19)	0.31*

* Chi^2 test, ** Wilcoxon test, *** t-test
CI = Confidence interval

We did, however, find a significant association between fibrinogen concentration after intervention and the risk of transfusion, but this effect was no longer significant after adjustment in the multivariable analysis (Table 3).

There was no difference in adverse events in the two groups at 24 hours post-intervention, including dizziness, shivering, headache, abdominal pain, nausea or vomiting. Furthermore, there were no thromboembolic complications in either group by the 6-week follow-up and very few readmissions, with no difference between the groups.

Table 3. Post hoc univariate and multivariate analysis for odds ratios of RBC transfusion. Intention to treat analysis. (*Adapted from "Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized control trial"*[1]*).*

Variable	Univariate analysis			Multivariate analysis		
	Odds ratio	95% CI	p-value	Odds ratio	95% CI	p-value
Fibrinogen level at 15 minutes	0.65	0.47-0.87	0.005	0.9	0.6-1.34	0.605
Centre No.2	0.38	0.12-1.07	0.08	0.66	0.16-2.67	0.561
Centre No.3	0.51	0.19-1.30	0.16	0.38	0.08-1.68	0.211
Centre No.4	0.68	0.32-1.52	0.34	0.6	0.19-1.96	0.391
Trauma	2.82	1.50-5.46	0.002	3.96	1.54-11.04	0.006
Tissue	2.11	1.07-4.45	0.04	2.17	0.82-6.3	0.133
Baseline estimated blood loss (ml)	3.56	1.88-6.96	<0.001	3.36	1.32-9.03	0.013
Baseline haemoglobin (g/L)	0.95	0.93-0.97	<0.001	0.38	0.24-0.58	< 0.005
Baseline crystalloids (L)	1.49	1.02-2.19	0.04	1.32	0.75-2.37	0.342
Crystalloids post-intervention (L)	1.62	1.14-2.31	0.007	1.97	1.21-3.38	0.009
Systolic blood pressure <100mmHg	2.60	1.03-6.28	0.04	0.62	0.14-2.91	0.535

CI = Confidence interval

Out of the 235 anaesthetists that evaluated blinding, a total of 220 (94%) had no idea about treatment allocation. However, nine (4%) guessed that their patient had been allocated to

fibrinogen due to the presence of foam in the tubes, and one knew their patient had been allocated to placebo due to deliberate un-blinding as a result of universal urticaria.

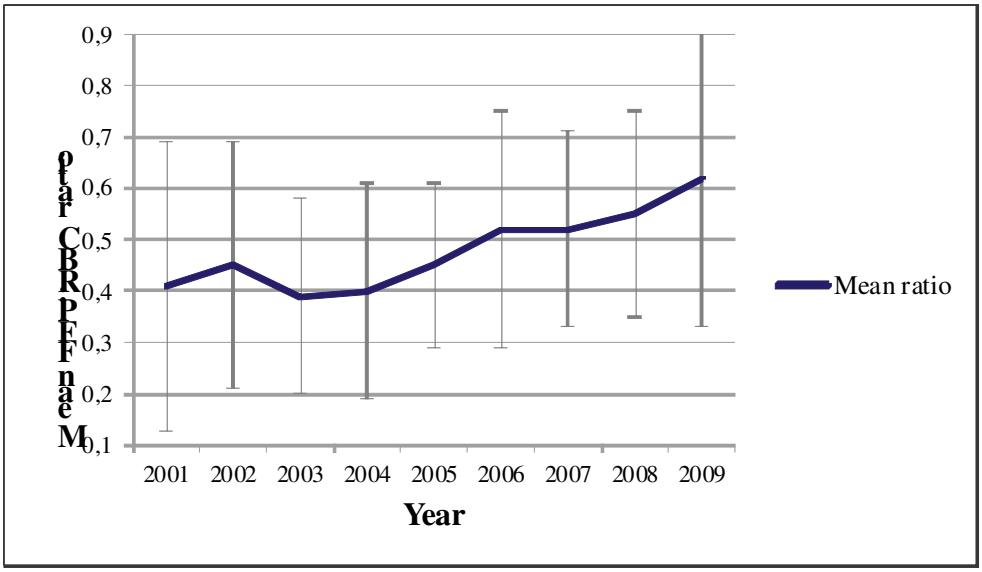
Out of the 1,967 women assessed for eligibility in the trial 22 (1.3%) fulfilled exclusion criteria, 592 (30%) declined to participate, and 449 (23%) were unable to give informed consent due to: 1) the acute situation (10%), 2) their psychological state (7%), 3) the language barrier (29%), 4) were uninformed of the study (48%), or 5) had other reasons (6%) (Figure 2). A total of 186 of the included women (76%) had had a positive experience of the trial, but 39 (16%) would have liked more information, 12 (5%) found timing of consent difficult, and 2 (1%) regretted participating in the trial.

Study II: Massive postpartum transfusion: a multidisciplinary observational study

A total of 245 women received massive transfusion of ≥10 units of RBC due to PPH, with 128 (52.2%) requiring hysterectomy in an effort to gain haemostasis. A total of 163 (66.5%) gave birth by caesarean section, 19 (7.8%) by instrumental delivery and 63 (25.7) by vaginal delivery. The median total blood loss was 8,000 ml ranging up to 53,000 ml, and with 57 women (24.2%) receiving more than 20 units of RBCs. The women spent a median of 9 days (IQR: 6-14) in hospital, with 170 (69.4%) spending a minimum of 24 hours in an Intensive Care Unit. Two women (0.8%) died, and an additional six (2.4%) had a cardiac arrest.

Haemorrhage started either just before or just after delivery (median 0 minutes, IQR: -7; +8 minutes), but first surgery after vaginal delivery was not performed before a median of 70 minutes (IQR: 41-157) after haemorrhage started. For all deliveries the median time from haemorrhage to the first RBC transfusion was 120 minutes (IQR: 49-229). The mean ratio of FFP:RBC given at the end of surgery leading to haemostasis was 0.45 (±0.23), with a significant increase from the 2001 to 2009, p=0.005 (Figure 3). The majority of RBC transfusions in women requiring hysterectomy were given before or during hysterectomy (median 13, IQR: 10-19).

Figure 3. Mean FFP:RBC ratio of all women from 2001 to 2009 .Whiskers indicating Interquartile range.

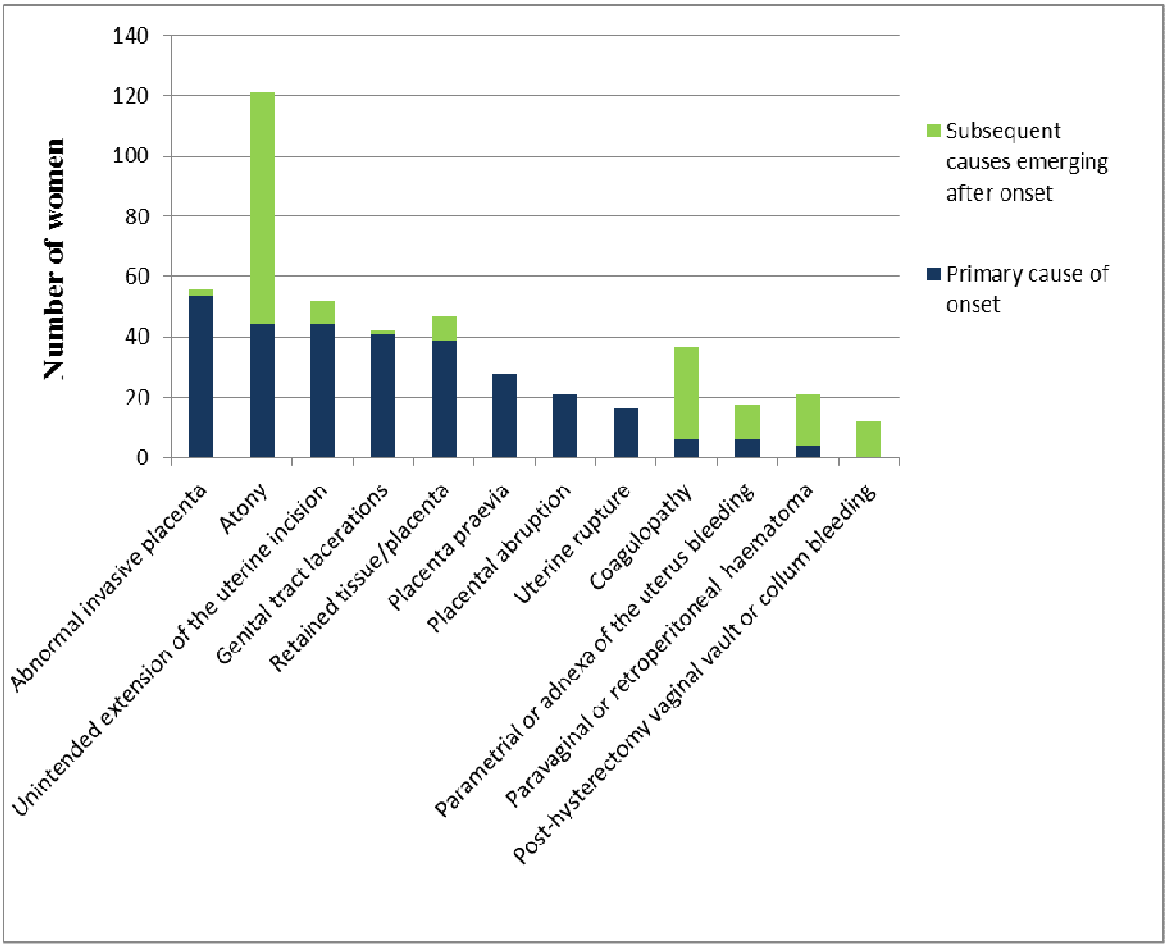


FFP = Fresh Frozen Plasma, RBC = Red Blood Cell.

From the data we had available, we identified 23 known risk factors seen either antenatally, during labour and delivery, or postpartum (Table 1). A total of 244 (99.6%) had at least one of these risk factors, 191 (78%) had an antenatal risk factor, 217 (89%) had a labour or delivery risk factor, and 80 (33%) had a postpartum risk factor.

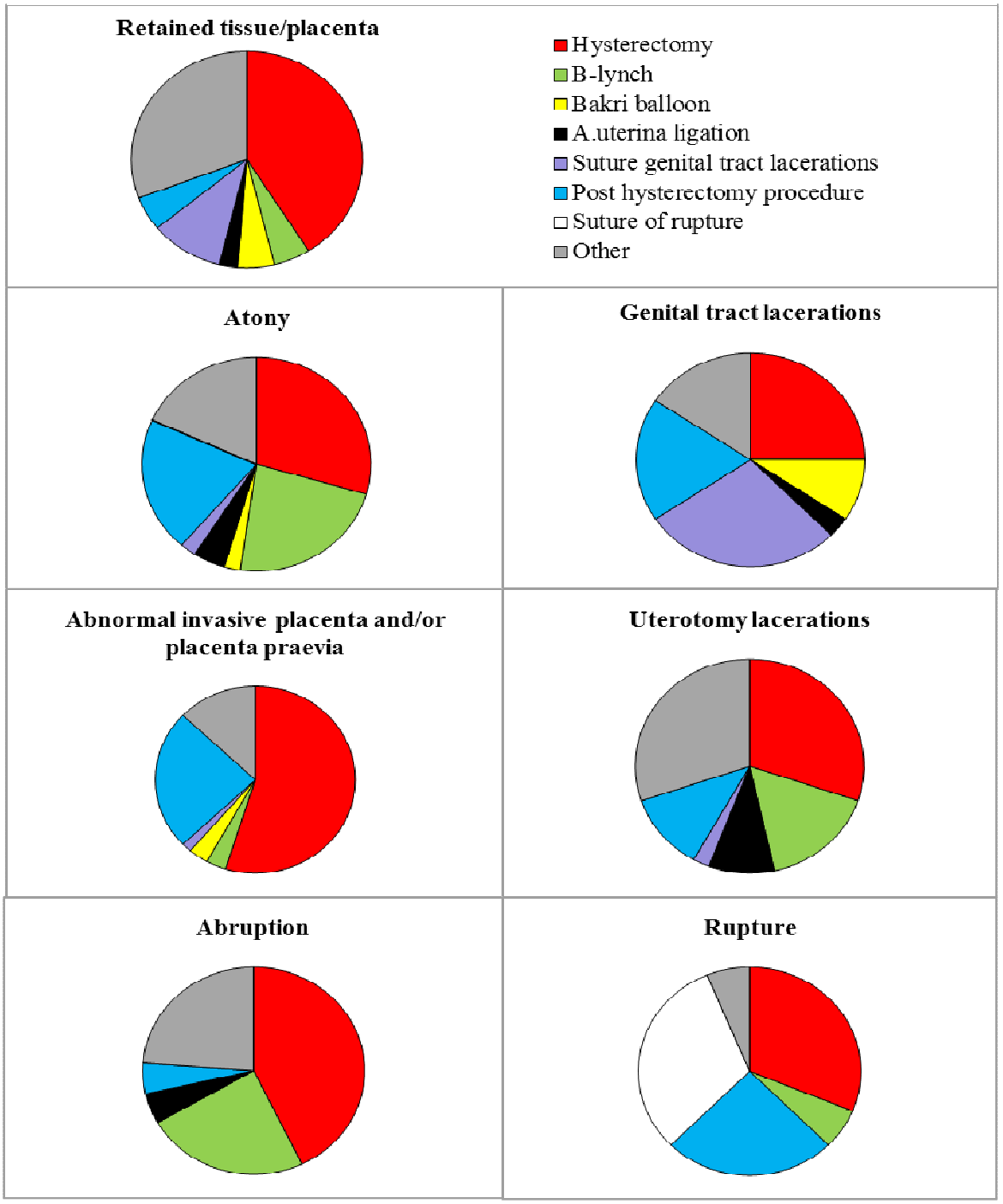
Causes of PPH were divided into causes of onset and subsequent causes not involved in the onset of PPH. There was a wide variation in causes overall and also a variation in the two subgroups of causes with the main causes of onset dominated by atony (n=93; 38%), abnormal invasive placenta (n=62; 25%), unintended extension of the uterine incision (n=59; 24%), genital tract lacerations (n=60; 24%), and retained tissue (n=42; 17%) and subsequent causes dominated by atony (n=77; 53%), coagulopathy (n=31; 21%) or haematomas (n=17; 12%) (Figure 4).

Figure 4. Total number of women with each cause divided into primary causes and subsequent causes emerging after onset of haemorrhage. Multiple causes were possible. (*Adapted from the manuscript "Massive postpartum transfusion: a multidisciplinary observational study"*[2])



In the 64 cases of AIP, only 6 were identified prior to delivery, all of which needed a hysterectomy. A total of 36 (56%) of women with an AIP had placenta praevia, 36 (56%) had a previous caesarean section of which 26 (41%) had both. Leaving a total of 18 women (28%) that had neither. However, the 36 women with placenta praevia and AIP constituted 86% of women with placenta praevia.

Figure 5. Procedures that gained haemostasis for each primary cause of onset.



A wide variation of procedures was performed in an attempt to gain haemostasis. Hysterectomy was performed most widespread (n=128, 52%), closely followed by suturing of genital tract lacerations (n=95, 39%), intrauterine palpation (n=85, 35%), extra suturing of the uterotomy (n=76, 31%), and B-lynch suture (n=71, 29%). There was a large variation in the procedures' ability to gain haemostasis, with 100% of splenectomies (n=4), 70% of hysterectomies (n=90), and 67% of embolizations (n=2) gaining haemostasis. The procedure that gained haemostasis varied between the different causes. Hysterectomy had the most substantial role in gaining haemostasis in cases of AIP and/or placenta praevia, placental abruption and retained tissue (Figure 5).

Table 4. Characteristics of women with massive postpartum transfusions with and without hysterectomy. Comparison by univariate logistic regression. Data presented as n (%), mean ± SD or median [IQR]. (*Adapted from the manuscript "Massive postpartum transfusion: a multidisciplinary observational study"*[2])

	Hysterectomy	No hysterectomy	p-value
Maternal characteristics			
Maternal age	34.0 ± 4.6	31.3 ± 5.1	**< 0.001**
Gestational age (missing n=12)	266 ± 23.9	274 ± 24.1	**0.012**
Parity	2.4 ± 1.2	1.6 ± 0.8	**< 0.001**
Previous caesarean section	50 (39.1)	24 (20.5)	**0.002**
Placenta praevia	36 (28.1)	6 (5.1)	**<0.001**
Emergency caesarean section	59 (46.1)	66 (56.4)	0.11
Birthweight >4,000g	15 (11.7)	29 (24.8)	**0.009**
Characteristics of PPH and treatment			
Number of RBCs before first PLT (missing n=62)	10 [6-13]	8 [5-10]	**0.006**
Number of FFP before first PLT (missing n=62)	4 [2-5]	2 [1-4]	**0.025**

RBC = Red Blood Cell, FFP = Fresh Frozen Plasma, PLT = Platelets. Bold indicates significant $p < 0.05$

We found that the 128 women requiring hysterectomy had a higher rate of previous caesarean section (p=0.002), placenta praevia (p<0.001), and AIP (p<0.001). Furthermore, they had greater blood loss (p<0.001) and received more units of RBCs (p<0.001), FFP (p<0.001) and PLTs (p<0.001) than women not requiring a hysterectomy. The FFP:RBC ratio was also higher in the

hysterectomy group at time of haemostasis (p=0.010), but they received significantly more RBCs before their first PLT transfusion (p=0.006) (Table 4).

A total of 38 (29.7%) of the hysterectomies performed did not lead to haemostasis. Women requiring further surgical management had a significantly higher rate of previous caesarean section and received higher volumes of RBCs, FFP, PLTs and colloids, and also had a higher ratio of FFP:RBC before initiating the surgery that led to haemostasis (Table 5).

Table 5. Comparison of characteristics of women requiring hysterectomy not leading to haemostasis with hysterectomy leading to hysterectomy. Data presented as n (%), mean ± SD or median [IQR].

	Hysterectomy, non-haemostasis, n= 38	**Hysterectomy, haemostasis, n=90**	**p-value**
Maternal characteristics			
Previous Caesarean section	21 (55.3)	29 (32.2)	**0.020***
Characteristics of PPH			
Time from start of life threatening haemorrhaging to haemostasis (hours:minutes)	11:52 [5:48-20:52]	3:00 [2:00-4:52]	**0.000****
Characteristics of transfusions			
Total no. of RBC at haemostasis	20 [15-27.5]	14 [11-19]	**0.000****
Total no. of FFP at haemostasis	10 [6-17.5]	7.5 [4-10]	**0.007****
Total no. of TRC at haemostasis	3.0 [1-3]	2.0 [0.75-3]	**0.035****
FFP:RBC ratio before start of haemostasis surgery	0.45±0.28	0.28±0.31	**0.007*****
Total crystalloids (ml)	8,000 [7,000-11,200]	6,000 [5,000-8,000]	**0.000****
Recombinant Factor VIIa, any	13 (36.1)	15 (17.4)	**0.034***
Complications			
Hospitalization (days)	11 [8-19]	9 [6-16]	**0.048****
Total blood loss (ml)	11,100 [9,000-14,000]	8,800 [7,500-11,000]	**0.003****
Time in intensive care (n=242) (days)	1 [0.75-2.5]	1 [0.5-1]	**0.006****

PPH = Postpartum haemorrhage, RBC = Red Blood Cell, FFP = Fresh Frozen Plasma, PLT = Platelets
*Chi^2 test, ** Kruskal-Wallis analysis, *** t-test. Bold indicates p <0.05

Study III: Causes and predictors of postpartum blood loss: a cohort study

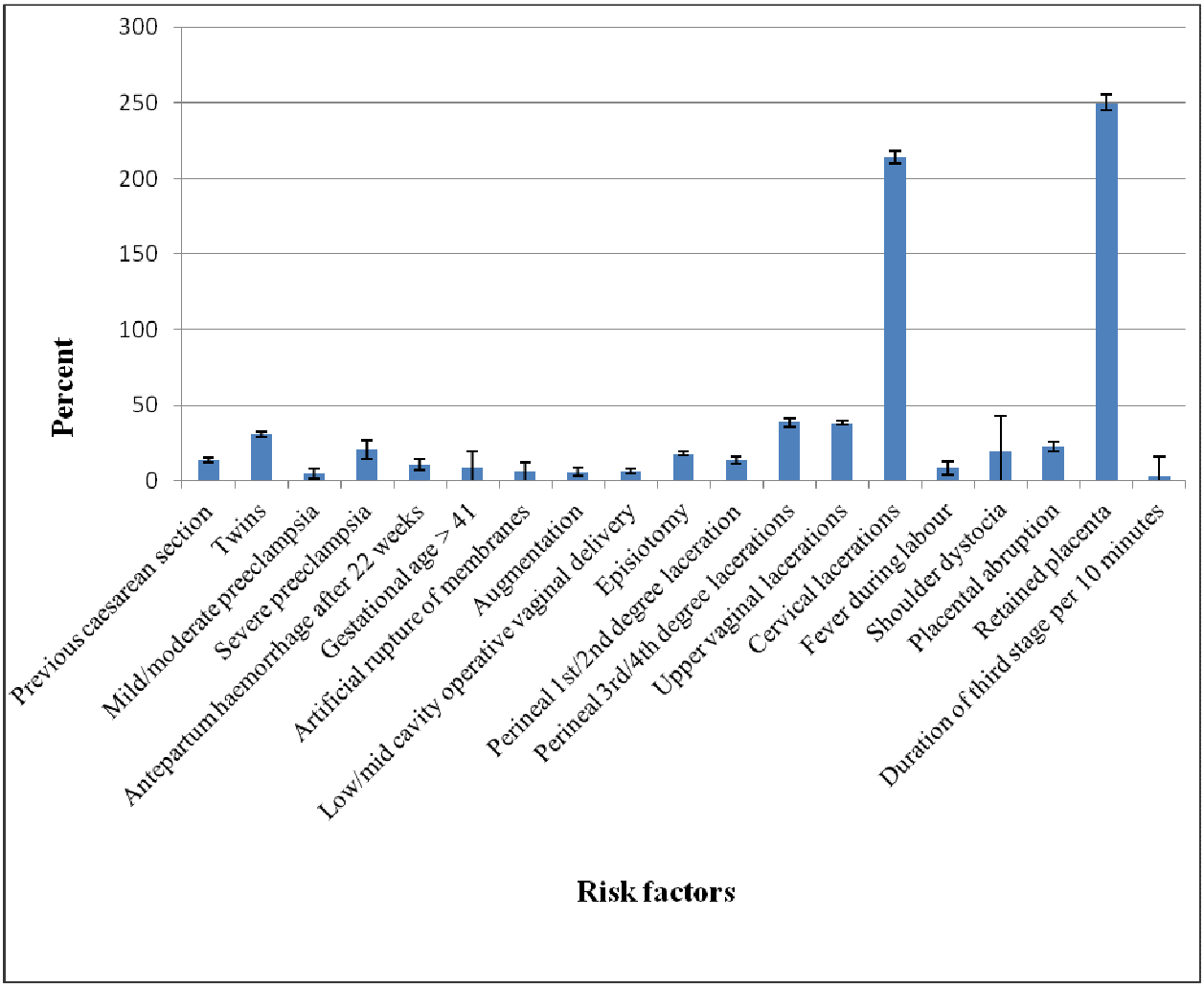
We identified 43,357 vaginal deliveries with a median of blood loss of 300 ml (IQR 200-400). There was a significant change in the distribution of causes the higher the cut-off used for defining PPH in the cohort. In cases of blood loss ≥500 ml (n=7,514) retained placenta accounted for 12%, lacerations 57%, and other causes including atony the remaining 31%. When increasing the cut-off to blood loss ≥1,000 ml (n=2,198) retained placenta accounted for 34%, lacerations 44%, and other causes including atony 22%. Further increase in the cut-off to blood loss ≥1,500 ml (n=1,113) led to retained placenta accounting for 47%, lacerations 37%, and other causes including atony 16%. Finally, in the cohort with a cut-off of blood loss at ≥2,000 ml (n=546) 53% were caused by retained placenta, 34% by lacerations, and 14% by other causes including atony.

A multivariate linear regression model was used to identify all available risk factors with a significant effect on the prediction of quantity of blood loss. This model accounted for 23.2% of the variability in quantity of postpartum blood loss (R^2=0.232).

Figure 6 represents the variables with the highest significant effect on prediction of quantity of postpartum blood loss in the final multivariate analysis model, illustrated in percent change. Uterine rupture, uterine inversion and eclampsia all had very high significant effects, but had very wide CIs as they consisted of less than five cases each, and have therefor not been included in the illustration. The effect of the duration of the third stage of labour was decreased substantially in the multivariate analysis compared to the univariate analysis, a reduction that was mainly facilitated by including "retained placenta" in the model (see full model analysis in the manuscript: *"Causes and predictors of postpartum blood loss: a cohort study"*[3]). Figure 6 illustrates the minimal effect the third stage of labour has on the predicted quantity of postpartum blood loss if a retained placenta is identified.

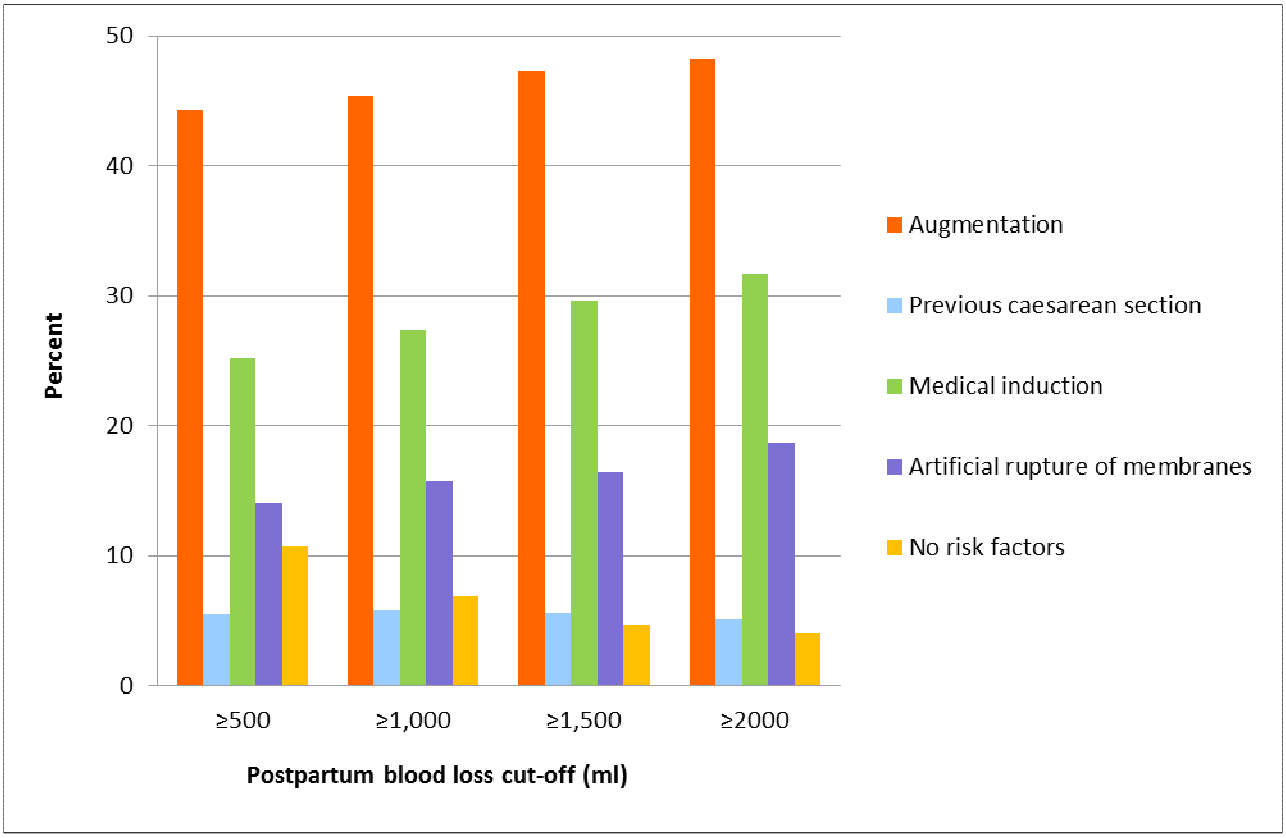
We also identified factors with a negative effect on prediction of quantity of postpartum blood loss, i.e. a protective effect on blood loss. These included second or third parity (-3.17%, CI: -4.3 to -1.8), oligohydramnios (-6.0%, CI:-10.7 to -1.1), chorioamnionitis (-29.7%, CI: -46.3 to -7.7), gestational age 22-32 weeks (-19.8%, CI:-24.1 to -15.1) and gestational age 33-36 weeks (-6.9%, CI: -10.3 to -3.6).

Figure 6. Percent change each risk factors affected the mean predicted quantity of blood loss. Whiskers represent Confidence Intervals.



Some of the identified risk factors are caused by procedures performed by obstetricians and midwives at a previous delivery or at the delivery in question (e.g. augmentation, previous caesarean section, medical induction or artificial rupture of membranes). These iatrogenic risk factors play an increasing role, the larger the cut-off used for defining PPH. Furthermore, fewer women had none of the risk factors included in the model, the larger the cut-off (Figure 7).

Figure 7. Percentage of women with iatrogenic risk factors or no risk factors for different definitions of postpartum blood loss.

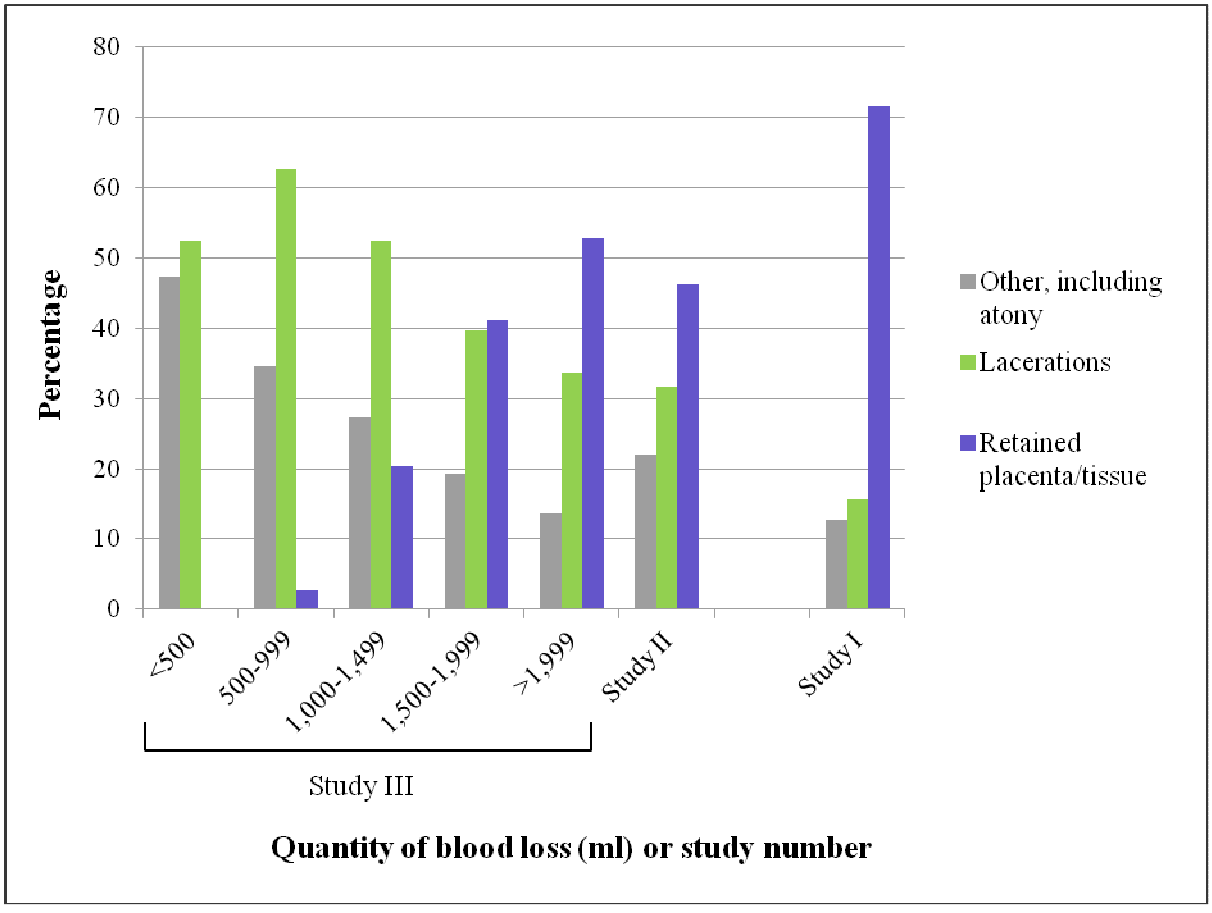


Women with "no risks" had none of the following risk factors: previous caesarean section, multiple pregnancy, hypertensive disorders, antepartum haemorrhage, oligohydramnios or chorioamnitis, gestational age <42 weeks, augmentation or induction of labour, low/mid cavity operative delivery, episiotomy, lacerations, fever during labour, uterine inversion, shoulder dystocia, epidural analgesia, uterine rupture, placental abruption and retained placenta.

Causes of PPH in vaginal deliveries: Study I, II and III

In all three studies, we identified the causes of PPH. Even though they are not all assessed in the same way, it is still interesting to compare them, as they represent three different severities of PPH. Study III consisted of vaginal deliveries, where each case was assigned a single cause. Therefore, we applied the same method to study I and II, including only vaginal deliveries and giving all women only one cause. "Retained placenta/tissue" was assigned first, then "lacerations" and finally "others, including atony" (see methods and materials). Study II and III are population based studies. As study II was comprised of the most severe cases of PPH it can be seen as a continuation of study III that mainly consisted of cases with PPH <2-3L (figure 8). Study I included a selected cohort of women able and willing to give informed consent, and is therefore not directly comparable to the population based studies, but it is still shown to the right in figure 8. Figure 8 illustrates the increasing role of a retained placenta and the decreasing role of atony the higher the blood loss in cases of PPH.

Figure 8. Distribution of causes for all vaginal deliveries in study I, II, and III.



Discussion

Overall findings

- Severe PPH could not be prevented with a fixed pre-emptive dose of fibrinogen in women with normofibrinogenaemia. The study was not large enough to evaluate rare complications such as thromboembolisms.
- Women with massive postpartum transfusion had a high incidence of severe morbidity and hysterectomy. Only 70% of the hysterectomies resulted in haemostasis. Women treated with hysterectomy had higher blood loss, and received more transfusions of RBCs, FFP and PLTs, than women not treated with hysterectomy during massive postpartum transfusion.
- The distribution of causes of PPH varied depending on the severity of PPH, with atony playing a smaller role and retained placenta a larger role than first anticipated. Retained placenta was, furthermore, a strong predictor of quantity of blood loss and diminished the effect a prolonged duration of the third stage of labour had on prediction of quantity of blood loss.

Strengths and limitations

Study I

Completing an RCT in an acute setting in obstetrics in the time frame planned, and only using independent funding is an accomplishment and strength in itself. What further strengthened the study were the successful block randomisation and allocation concealment reducing confounders and selection bias, and double-blinding of the majority of clinicians and all patients, thereby limiting performance bias. Furthermore, the external validation is strengthened through our ability to include women, through a multicentre set-up with few exclusion criteria. Limitations in this study are, however, also present. Considerably fewer women included in the trial received RBC transfusions than first anticipated (placebo group 21.5%, estimate for sample size calculations 57%). The fact that we did not meet a transfusion rate of 57% results in a lower statistical power. If we wanted to find a risk reduction of 33% (and 15% dropout) we would have needed to include 1,021 women. Therefore the study was not powered to evaluate the planned effect of fibrinogen concentrate. In a recent randomised controlled trial of 56 women with severe PPH >1,000 ml, 31 women (55%) received at least one blood transfusion, however, they also

found no risk reduction when increasing the fibrinogen level from approximately 3 g/l to 4 g/L. One of the reasons for our low rate of transfusions could be the inability to include women with the most severe PPH, either due to the women being incapable of giving an informed consent or due to the clinicians being unable to cope with further challenges in an already critical situation. This is discussed further below (p.42). We have as yet not been able to extract data regarding women who were not included in the study with regard to their quantity of PPH. Another reflection of the low inclusion rate of the most severe cases of PPH, can be seen in the low rate of women with fibrinogen concentrate <2 g/L at inclusion, which was the group expected to have the highest effect of an increase in fibrinogen. Even four hours after intervention when the estimated blood loss was close to 3 L, the mean fibrinogen level in the placebo group was above 4 g/L. Our findings of a higher level of fibrinogen in women with severe PPH are discussed below (p.43).

Even though 120 women received 2g of fibrinogen concentrate it was not enough to assess the risk of thromboembolic complications. In our population the risk of venous thromboembolic complications is estimated at 0.7-2.0/1,000 pregnancies[63], therefore even if fibrinogen concentrate caused a two-fold increase in risk, we would not necessarily have seen a single case in our cohort during the six week follow-up.

Study II

With massive transfusion being a rare occurrence in obstetrics, a cohort of this size, with only six case files missing, gives a good description of a group of extremely severe cases. Identification of the cohort through Danish national registries representing the majority of births in Denmark in the chosen period increased the external validity. Detailed validation of the data obtained through patient files using standardised abstraction forms for all patients, excluding women receiving massive transfusion due to non-obstetric complications, strengthened the internal validity, and by using only one abstractor we minimised interrater variability. Some aspects of selection bias have been accommodated through well-defined inclusion criteria of ≥10 RBC transfusions and childbirth, establishing a cohort from national registries of known high validity[105,119]. However, selection bias in general is one of the major limitations of this study, due to non-random assignment of not only treatment, but in our case probably also the outcome of hysterectomy, both of which could be highly influenced by confounders, that we could not take into consideration. These confounders include experience level of the clinicians involved in treating these severe cases and availability of blood products. Both of these correlate to some extent with a large birth place. Size of birth place was included in the analyses, but this can in no

means take the whole effect into account. Multivariate analysis increases selection bias further, excluding all cases with missing values in variables included, perhaps causing a selection of more severe cases where recording of information was more thorough. All in all determination of causation is not possible due to confounding-by-indication where the decision to perform hysterectomy could be influenced by failure of treatments attempted before the decision to perform hysterectomy. Furthermore, our findings could be due to random error, as the level of significance at $p<0.05$ still leaves a risk of 1/20 that our findings could be obtained by chance.

Study III

Cohort studies of this size increase the probability of identifying true association, and by using multivariate regression analysis we were able to account for multiple factors that could influence the prediction of quantity of postpartum blood loss. Known high validity of the Obstetric Database strengthens this study [107], and as the population included has known homogeneity with the rest of Denmark, external validity is increased[120]. The limited exclusion criteria reduced selection bias, but could not eliminate this completely due to exclusion of cases with missing data. In addition missing data contributed to further limitations, as we were not able to include possible confounders (pervious PPH, BMI and birthweight) in the model. As in study II there is a risk of random error, albeit smaller as the majority of p-values fall below 0.001.

Clinical trials in emergency obstetrics

Clinical trials are important in all fields of medicine, but are fairly rare in emergency obstetrics probably due to challenges with informed consent and the recruitment process[111,113]. We also met several of these challenges and had an initial slow inclusion rate in study I, partly due to the multi-centre set-up and 24 hour recruitment under emergency situations. Under these circumstances it is not possible for the project coordinator to be on site for each inclusion but relies instead on the staff on duty feeling properly prepared for all aspects of the inclusion process. It is well known that slow recruitment is one of the main problems in clinical trials, with as little as 31% of trials meeting recruitment targets, even fewer in emergency medicine[121,122]. We managed to overcome the slow inclusion rate once all fours sites had got used to inclusion, but the trial was still affected by some of the known obstacles of trials in emergency obstetrics, as illustrated by the large number of women declining to participate (30%) or unable to give informed consent (23%). This could be the reason that we did not include women with the most severe PPH, thereby not meeting our transfusion rates used for the sample size calculation.

Recent qualitative studies have identified areas related to the organisation, the staff recruiting, and the process of informed consent as the major obstacles[121–125]. Organisational barriers include the setup of the trial and the incentives of the organisation to focus on research[122,124]. Although we recruited from university affiliated hospitals, we felt that some of the centres were less used to being involved in clinical trials, and some of the staff expressed that they were not hired to do research. The ability for an organisation to inform staff and patients about research taking place can play a role in both staff' and patients' attitudes to research. On the homepages of the four hospitals in our study only one (Nordsjaellands Hospital) has direct information regarding research on their front page. In comparison Yale and Harvard affiliated hospitals have information regarding research trials being conducted there, on the front page of their home pages (http://www.ynhh.org and http://www.massgeneral.org/). Other obstacles influencing the recruiting staff involve conflicts in regard to time available, feeling the need to prioritise the clinical side of work at the expense of research, or not wanting to approach vulnerable patients[122,125]. An improvement of these factors could enhance the ability to include patients during an emergency setting, e.g women with severe PPH.

As mentioned before informed consent is requested by law, and requires not only that a patient can receive information and understand it, but also that someone is capable of giving sufficient information[110]. The fact that 23% were unable to give informed consent in our trial could possibly reflect not only difficulties receiving information, but also difficulties providing information in the acute situation. In a recent study enrolling patients postpartum at time of diagnosis of a retained placenta, staff expressed they had limited time to give information and often ended up giving very simplified information[125]. The women recruited to the trial expressed gratitude to receiving sparse information at a point in time when they felt anxious and exhausted. However, in hindsight, the majority felt that they had not given a fully informed consent and would have preferred more information antenatally. The staff on the other hand found that it might scare women if all women were to be informed of the study, where only 2% would become possible candidates, even though this pathway is recommended by the RCOG[126]. Our study had tried to give summary information antenatally, but failure to fulfil this can be seen in the fact that 48% of women declining to participate felt uninformed on the study and 16% of included women would have like more information. If antenatal information had been successful, this too could have improved our ability to include women with the most severe PPH.

Coagulopathy in PPH

Coagulopathy was found in 35% of cases in study II, some of which had not been recognised at the time of events. This together with the large proportion of hysterectomies not resulting in haemostasis, supports the notion that coagulopathy is not always identified or considered during the attempt to gain haemostasis through surgery alone[4,23,73]. Even though the transfusion rate used in our sample size calculation was not met in study I, we found no significant effect of early pre-emptive fibrinogen as a strategy for preventing development of a more severe PPH due to coagulopathy. Therefore, perhaps we should focus on viscoelastic assays early on to give a more goal directed treatment. Several studies have recognised the potentials of viscoelastic assays in identifying coagulopathy in women with PPH[64,127], but not all delivery units have access to these assays, and can we reject the effect of pre-emptive fibrinogen for all cases of PPH based on one single trial? – probably not. As mentioned before the transfusion rate in study I was lower than anticipated, and the included women had higher levels of fibrinogen than expected in relation to the study by Charbit et al[84]. This could indicate that the women in our trial had less severe bleeding and perhaps received smaller volumes of crystalloids and colloids than the study population of Charbit et al, where no data on blood loss or crystalloid/colloid infusion is available. We found a decreased risk of transfusion in the group with an increased fibrinogen concentration after intervention in the univariate analysis, which corresponds with the findings from Charbit et al. However, after including blood loss, crystalloids, colloids etc. in our multivariate analysis, the difference was no longer significant (Table 3). This leads us to question whether the findings from Charbit et al would have been significant if they had done the same, or whether the lower fibrinogen concentration simply was a result of more severe PPH and dilution, as identified in a similar study[128]. We know that coagulopathy develops due to both dilution and consumption[66], and a low fibrinogen concentration is a sign of coagulopathy. However, we do not know at which specific level a low fibrinogen critically impairs coagulation leading to further blood loss in women postpartum. A Cochrane study found it possible that fibrinogen concentrate reduces the need for blood transfusions, but included studies investigating both pre-emptive treatment and treatment of known or suspected hypofibrinogenaemia in various fields of medicine[129]. Studies measuring antenatal fibrinogen levels have shown diverging results[128,130], but hopefully future trials will demonstrate whether women with more severe PPH could benefit from pre-emptive fibrinogen substitution[89], or whether substitution should be based on viscoelastic assays where the effect of dilution is taken into account[88]. So far a pilot study investigating the effect of fibrinogen concentrate in women with PPH and low levels of

fibrinogen (measured by viscoelastic assay "FIBTEM"), showed no reduction in the need for blood transfusion[131].

There are some circumstances where there is no time to wait for analyses, but where focus instead should be on immediate life-saving measures. This includes profuse uncontrollable PPH, where substitution with RBCs in inevitable, cases similar to the population in study II requiring ≥10 units of RBCs. In this cohort we found that women not requiring hysterectomy received their first PLT transfusion after fewer RBC transfusions, but received a lower FFP:RBC ratio at time of haemostasis than women requiring a hysterectomy. Identifying risk factors is a complex matter in a study like this, as we cannot tell the difference between factors that were a consequence of hysterectomy, and factors that influenced the decision to perform hysterectomy. We know that women treated with hysterectomy required more transfusions, longer time in surgery and longer hospitalisation, but we also know that they bled more and perhaps this led to the decision of hysterectomy and additional blood transfusions. Likewise a higher FFP:RBC ratio could be due to a longer time in surgery, giving the anaesthetist time to consider a more balanced transfusion. However, the treatment with PLTs is more complex. Transfusion of first PLT after fewer RBCs in women requiring hysterectomy could be because there was no profuse bleeding and the clinicians were less stressed and had time to consider platelets (which is the opposite of our considerations regarding FFP), or that women receiving more RBCs before their first PLTs – and thereby more dilution –bled more profusely due to coagulopathy, which in turn led to hysterectomy. The only other study investigating the effect of PLT transfusions in PPH concluded in their retrospective study, that the 12 women receiving PLT transfusions had either antenatal thrombocytopenia, placental abruption or blood loss >5,000 ml, and there was therefore no need for early fixed-ratio transfusion of PLTs[132]. This conclusion was based on the fact that PLT transfusion was only used in these cases and therefore not necessary in other cases. However, without comparison on outcome in these two groups, it is difficult to conclude whether sufficient women received PLT transfusions. A study using an in-vitro model found that a fixed rate of PLT:FFP:RBC of 1:1:1 in postpartum women was associated with decreased coagulopathy measured by viscoelastic assay[133], and studies of massive transfusion in other populations have shown increased survival with a fixed rate of early PLT transfusions, thereby supporting the use of fixed ratios[78,134,135].

We found no benefit of a high FFP:RBC ratio in study II, but other studies on severe PPH have reported a reduction in the need for interventional procedures and an increase in the success rate of hysterectomy compared to lower ratios[94,99]. The majority of studies of massive transfusion due to PPH have, however, not compared ratios between groups, but only stated the overall ratio in their cohorts ranging from 0.3 to 0.8[136–139]. The overall mean FFP:RBC ratio in study II of 0.48 is within this range. The FFP:RBC ratios in the more recent years of study II (2008-2009) had higher ratios, probably due to the increased focus on balanced transfusions and introduction of a transfusion protocol in 2007 in Denmark[140].

Predicting the few and treating the cause

In study III risk factors affecting the prediction of postpartum blood loss were identified in a multivariate linear regression model. However, we also saw that only 11% of women with blood loss ≥500 ml, and 5% of women with blood loss ≥1,000 ml, had none of the identified risk factors. Likewise in study II we found only one patient (0.04%) requiring massive transfusion that had no risk factors for PPH. The majority of risk factors identified in the two studies were the same, but study II also included caesarean section deliveries, cases of placenta praevia, previous PPH, age >35 years, and BMI >35 as risk factors. Study III included extra data regarding fever during labour, amniotic fluid abnormalities, uterus inversion, shoulder dystocia and epidural analgesia as risk factors, making the studies less comparable. Despite of this, it seems evident that the vast majority of most severe cases of PPH have at least 1 risk factor. This is in line with a recent study of women with PPH requiring ≥8 RBC transfusions, where 3% (5/181) did not have any risk factors[141]. Other studies have shown a higher proportion of cases without risk factors e.g. 30% of cases with PPH and transfusion[49], and 61% of cases with PPH ≥500 ml, probably due to a variation in included risk factors. As noted previously many of the identified risk factors have a small effect on predicted blood loss and are also seen in women not developing excessive blood loss. However, risk factors should not be ignored, but instead help the clinician take extra precaution, even in cases with one or two low effect risks.

Up until now, prolonged duration of the third stage of labour has been considered one of the risk factors that has a high effect on postpartum blood loss[26,27,29,47]. In study III we found that the majority of this effect was due to a retained placenta, including cases requiring manual removal and cases of AIP. This is quite controversial, as the main focus in recent years has been on early manual removal of placenta irrespective of whether there is ongoing haemorrhage[26,27,31]. In

contrast our results suggest that if there is no retained tissue or the need for manual removal, then a prolonged duration of the third stage of labour will not lead to excessive blood loss. This approach requires early identification of a retained placenta, a condition recognised in all three of our studies and similar studies as a major cause of PPH[12,42,43]. To date this has proven difficult, with only up to 50% of the most severe cases being identified prior to delivery[37,142,143].

Retained placenta and other causes of PPH including lacerations and uterine atony, were found to change in distribution the larger the quantity of blood loss in study III. Especially atony seemed to play a much smaller role than the traditionally ascertained 70%[21]. Study III and Figure 8 comparing the three studies has probably underestimated the role of atony due to cases only being assigned this cause as an exclusion diagnosis in the absence of lacerations or retained placenta. This change in distribution could also be due to an effect on atony of early administration of uterotonics, that is advised in the Danish guideline[82]. This could lead to progression of PPH for cases where uterotonics have no effect.
The original data for study I and II were assigned causes after review of patient charts, with study II dividing the causes further into causes of onset and causes arising after onset. Very few other studies have included secondary atony in their analysis, but it is described in case reports, and one study found that 27% of cases treated with internal iliac ligation due to PPH had secondary atony[4,144]. This is very close to our findings, perhaps suggesting that this is an underestimated cause in many cases of PPH. It is difficult to determine the physiology behind secondary atony. We know that haemostasis after placental delivery is obtained through both contraction and coagulation[55,62], then perhaps if one of these factors is insufficient the effect of the other is reduced. For example if coagulopathy develops due to ongoing haemorrhage then the placental site loses its' coagulative ability, causing blood to gradually fill the uterine cavity until the uterus finally cannot keep up due to exhaustion, and atony arises. In comparison atony arising after sufficient coagulation at the placental site might cause minimal haemorrhage.

Identifying the cause of haemorrhage is of great importance, as treatment strategies are different for different causes. Not only is it now obvious that coagulopathy cannot be treated with hysterectomy, but extensive lacerations in the genital tract including cervix also have limited effect of a hysterectomy[4]. But these causes can be overseen if focus is primarily on atony already from the start of PPH, due to an exaggerated role of atony in traditional literature.

Conclusion

For normofibrinogenaemic patients with severe PPH, pre-emptive treatment with 2 grams of fibrinogen concentration showed no effect on the need of RBC transfusions postpartum. There were no thromboembolic complications due to fibrinogen concentrate, but the cohort was not of sufficient size to evaluate this.

In more severe cases of PPH, where women were treated with massive transfusion the morbidity and rate of hysterectomy were high. Women treated with hysterectomy had higher blood loss, more transfusions of RBCs, FFP and PLTs, more RBC transfusions before initiating PLT transfusion, and longer hospitalisation than women not treated with hysterectomy. Furthermore, only 70% of the hysterectomies actually resulted in haemostasis.

The distribution of causes varied significantly depending on the cut-off used to define PPH, with atony playing a smaller role than first anticipated. A retained placenta played an increasing role the higher the cut-off used. The predictive effect of a prolonged duration of the third stage of labour on quantity of blood loss was diminished by the influence of a retained placenta.

Perspectives

The results of our studies contribute with findings regarding prevention and treatment of severe PPH, thereby providing new evidence for guidelines and new paths to follow in future research, so that we can avoid some of the severe cases of PPH, that are associated with such high morbidity.

Our experiences with informed consent in the clinical trial have the potential of helping others if steps are taken to improve focus on research from an organisational level. This could involve easy accessible information on websites or in waiting rooms regarding ongoing trials, benefits of research, and results of previous trials that have influenced treatment regimes. Hopefully this could help prepare patients for the possibility of being confronted with research protocols during an emergency situation, and giving women that want to know more, the possibility of access to in depth information. Staff should also be informed of the importance of research trials taking place, and can perhaps become more comfortable with enrolment strategies if all staff involved are prepared and support each other in these often hectic situations.

The absent effect of pre-emptive fibrinogen in our study will hopefully change some of the guidelines already existing that recommend fibrinogen concentrate if PPH exceeds approximately 1500 ml[145–147]. Not only did we not find any benefits of this treatment, but the safety in relation to thromboembolic complications has not been clarified. Therefore, treatment should be given with precaution and only in cases where the benefits have been proven. Today the use of fibrinogen for PPH is the main focus for several ongoing trials with the most recent taking our results into account[88,89]. New trials together with our findings of coagulopathy in the most severe cases of PPH can hopefully draw further attention to the role of coagulopathy and the importance of early recognition and treatment before it becomes incontrollable. This could in turn lead to fewer hysterectomies becoming necessary, and not being performed in cases of coagulopathy where removal will not lead to haemostasis.

For cases of profuse bleeding, where actions need to be taken immediately, our findings support the benefits of an early platelet transfusion. Training in transfusion algorithms alongside practice drills for PPH could benefit not only patients, but also obstetricians and anaesthetists by improving their collaboration in these emergency situations.

Our findings of the decreased role of atony in the onset of PPH should emphasise the need for early identification of other causes, perhaps including quick transfer to an operating theatre, to facilitate thorough investigation for retained tissue or cervix lacerations. Treatment of atony should, however, not be forgotten as PPH can have multiple causes not necessarily arising simultaneously. Furthermore, if identification of the majority of cases of retained placenta is deemed possible in the future, then sufficient precautions can be made to prevent excessive blood loss, focusing less on the duration of the third stage of labour and more on immediate manual removal of a known retained placenta. In addition, the majority of women suffering from PPH were found to present with at least one risk factor before or during labour, that together with the identified effects of each risk factor can guide the clinician on where to prepare for PPH.

Finally, we found that the distribution of causes varied depending on the cut-off used to define PPH, illustrating that studies regarding PPH are not always comparable. Perhaps the question is not what definition to use, but whether we need a definition of PPH. It is practical in a clinical setting to have a definition of PPH that can be used to trigger treatment strategies. In our large

cohort study 83% of all vaginal births lost less than 500 ml of blood. The traditional cut-off of 500 ml seems therefore relevant for initiating investigation of the causes, even though estimates in this range are known to be somewhat inaccurate[24]. A cut-off for quantity of blood loss or blood transfusions for defining severe PPH in order to initiate certain treatment strategies seems less warranted. Treatment of severe PPH should be based on individual observations, taking not only blood loss, cause of PPH, and dilution into account but also measurements of coagulation and vital functions. A definition of severe PPH could, however, be based on the need for transferral to an operating theatre, as this is known to have a major impact on women's experience during childbirth[125]. The definition or diagnosis could then be used to make sure these women were given extra attention postpartum. Definitions are, however, vital for research purposes where comparisons of studies or systematic reviews depend on uniform definitions. Definitions for research purposes could be obtained through Delphi procedures, where consensus is achieved from a group of experts, for example through the INOSS collaboration[148,149].

Future studies

The role of fibrinogen concentrate for prevention of development of severe PPH in women with hypofibrinogenaemia has yet to be investigated.

With regards to PPH requiring massive transfusion, the ideal study would be an RCT evaluating different transfusion protocols. Due to the scarce incidence of these cases an RCT would have to involve a multinational collaboration and would be affected by challenges in gaining informed consent. Taking this into consideration a larger observational study might be more feasible and would perhaps give us answers to some of our questions.

An RCT investigating the benefits of transfusion guided by viscoelastic assays is on the other hand feasible in an obstetric population with less severe PPH, and could perhaps lead to prevention of haemorrhage due to coagulopathy.

We need studies investigating early identification of retained placenta. These studies should not only focus on ultrasound modalities but should also consider investigating biomarkers involved in the placental development.

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Appendix

Paper 1: Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized controlled trial.

Paper 2: Massive postpartum transfusion: a multidisciplinary observational study.

Paper 3: Causes and predictors of postpartum blood loss: a cohort study.

BJA Advance Access published January 13, 2015

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doi:10.1093/bja/aeu444

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Pre-emptive treatment with fibrinogen concentrate for postpartum haemorrhage: randomized controlled trial

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Editor's key points

- Low fibrinogen is associated with excessive bleeding in postpartum haemorrhage.
- The effect of early empirical administration of fibrinogen concentrate on blood transfusion in postpartum haemorrhage was studied.
- In a multicentre, randomized trial of 249 subjects, pre-emptive administration of fibrinogen concentrate did not reduce red blood cell transfusion.

Background. In early postpartum haemorrhage (PPH), a low concentration of fibrinogen is associated with excessive subsequent bleeding and blood transfusion. We hypothesized that pre-emptive treatment with fibrinogen concentrate reduces the need for red blood cell (RBC) transfusion in patients with PPH.

Methods. In this investigator-initiated, multicentre, double-blinded, parallel randomized controlled trial, we assigned subjects with severe PPH to a single dose of fibrinogen concentrate or placebo (saline). A dose of 2 g or equivalent was given to all subjects independent of body weight and the fibrinogen concentration at inclusion. The primary outcome was RBC transfusion up to 6 weeks postpartum. Secondary outcomes were total blood loss, total amount of blood transfused, occurrence of rebleeding, haemoglobin <58 g litre^{-1}, RBC transfusion within 4 h, 24 h, and 7 days, and as a composite outcome of 'severe PPH', defined as a decrease in haemoglobin of >40 g litre^{-1}, transfusion of at least 4 units of RBCs, haemostatic intervention (angiographic embolization, surgical arterial ligation, or hysterectomy), or maternal death.

Results. Of the 249 randomized subjects, 123 of 124 in the fibrinogen group and 121 of 125 in the placebo group were included in the intention-to-treat analysis. At inclusion the subjects had severe PPH, with a mean blood loss of 1459 (SD 476) ml and a mean fibrinogen concentration of 4.5 (SD 1.2) g litre^{-1}. The intervention group received a mean dose of 26 mg kg^{-1} fibrinogen concentrate, thereby significantly increasing fibrinogen concentration compared with placebo by 0.40 g litre^{-1} (95% confidence interval, 0.15–0.65; $P=0.002$). Postpartum blood transfusion occurred in 25 (20%) of the fibrinogen group and 26 (22%) of the placebo group (relative risk, 0.95; 95% confidence interval, 0.58–1.54; $P=0.88$). We found no difference in any predefined secondary outcomes, per-protocol analyses, or adjusted analyses. No thromboembolic events were detected.

Conclusions. We found no evidence for the use of 2 g fibrinogen concentrate as pre-emptive treatment for severe PPH in patients with normofibrinogenaemia.

Clinical trial registration. ClinicalTrials.gov: http://clinicaltrials.gov/show/NCT01359878. Published protocol: http://www.trialsjournal.com/content/pdf/1745-6215-13-110.pdf.

Keywords: blood coagulation; erythrocyte transfusion; fibrinogen; postpartum haemorrhage

Accepted for publication: 5 October 2014

Postpartum haemorrhage (PPH) remains a leading cause of maternal morbidity and mortality.[1] Fibrinogen is an essential endogenous component of haemostasis,[2] and its plasma concentration increases during pregnancy.[3] Observational studies of patients with PPH indicate that a low fibrinogen concentration in the early phase of PPH is associated with excessive subsequent bleeding and blood transfusion.[4–7] Fibrinogen concentrate is widely used to correct acquired hypofibrinogenaemia, but evidence is lacking regarding the efficacy of this treatment.[8 9]

We aimed to assess the efficacy of a pre-emptive treatment with fibrinogen concentrate in patients with PPH,[10] and hypothesized that treatment with fibrinogen concentrate reduces the need for red blood cell (RBC) transfusion up to 6 weeks postpartum.

Methods

Ethical approval

The Danish National Committee on health research ethics approved this trial (protocol number 1002168/H-3-2010-004). Written informed consent was obtained from all subjects entering the trial.

Randomization and masking

This trial was investigator-initiated, without any financial or academic involvement from the manufacturers of fibrinogen concentrate. We designed a multicentre, double-blinded, centre-stratified trial with 1:1 parallel groups and used computer-generated allocation and blocks-of-four randomization (done by third party before study commencement). This was concealed by envelopes. When including a subject, an envelope was collected and the content decided/randomized the subject to either intervention or placebo. The stratification by centres implies that within each centre the number of envelopes with intervention or placebo was balanced, and an excess of envelopes was available at all times. We randomly assigned patients with primary PPH regardless of mode of delivery to early pre-emptive treatment with a single i.v. dose of fibrinogen concentrate (RiaSTAP©; CSL Behring, Marburg, Germany) or placebo (isotonic saline) administered by the anaesthetist on arrival in the operating theatre. Subjects received written project information during pregnancy. They were invited to participate and informed consent was sought by the attending anaesthetist at the pre-anaesthetic evaluation before Caesarean section, when labour epidural was performed, or following vaginal delivery with bleeding. In order to secure blinding, we used an anaesthetist doctor or nurse not involved in the treatment of the patient to carry out the randomization and dispensation. Subjects, care providers, outcome assessors, and the statistician were blinded to allocation. Opaque syringes (yellow coloured) were used to disguise the content of the study infusion. We assessed physician and subject blinding. No interim analysis was performed.

The FIB-PPH (FIBrinogen concentrate as initial treatment for PostPartum Haemorrhage) trial was conducted between May 30, 2011 and July 11, 2013 in four university-affiliated public tertiary care hospitals in the Capital Region of Copenhagen, Denmark (Rigshospitalet, Hvidovre Hospital, Herlev Hospital, and Hillerød Hospital). Each centre had 3500–7000 deliveries yr^{-1}, which represents 99% of all deliveries in this region (1% are home deliveries). The trial protocol, including planned statistical analyses, has been published elsewhere.[10]

Subjects

All subjects entering the trial were randomized once the following PPH inclusion criteria were met: (i) age $\geq$18 yr; and (ii) PPH defined as bleeding from uterus and/or birth canal within 24 h postpartum. Additional inclusion criteria were Caesarean section with an estimated perioperative blood loss >1 litre or vaginal delivery with either estimated blood loss >0.5 litre and intended manual removal of placenta or estimated blood loss >1 litre and intended manual exploration of the uterus because of continuous bleeding after delivery of the placenta (Fig. 1). All hospitals in the Capital Region of Denmark follow our national guidelines for treatment of PPH, including treatment with oxytocin. Blood loss up to 0.5 litre was assessed by visual inspection of the absorbent delivery pad, and thereafter determined by weighing of drapes and pads as described in the national guidelines.[11] Exclusion criteria were as follows: (i) known inherited coagulation deficiencies; (ii) antenatal anti-thrombotic treatment; (iii) pre-pregnancy weight <45 kg; or (iv) refusal to receive blood transfusion.

Intervention and placebo

The fibrinogen group received a fixed dose of 2 g of fibrinogen concentrate dispensed in 100 ml sterile water. The placebo group received 100 ml of isotonic saline. This was applied in a pragmatic early pre-emptive treatment regimen, giving a fixed dose to all subjects irrespective of body weight and not guided by measurement of fibrinogen concentration or other haemostatic measures. The solution was dispensed using syringe-pump infusion over 20 min according to the manufacturer's recommendation.[12] Additional interventions, including the use of tranexamic acid and i.v. fluids, were at the discretion of the attending physician supported by regional guidelines.[13] The anaesthetists following study drug administration and subjects at the 6 week follow-up were asked to evaluate the blinding.

Haemostatic monitoring and adverse events

Blood samples were taken before intervention (baseline) and 15 min, 4 h, and 24 h after infusion of the study drug. Specially trained nurses obtained the samples and assessed fluid, transfusion status, and adverse events at 4 and 24 h after intervention. The haemostatic monitoring has been described previously.[10] Data on blood transfusions were retrieved from charts and validated in the Blood Bank Database. We approached all subjects 6 weeks postpartum by telephone and asked about contact with general physicians or hospitals. Charts were assessed for readmissions and thromboembolic events.

Outcomes

The primary outcome was RBC transfusion during a 6 week follow-up period postpartum. Danish national guidelines

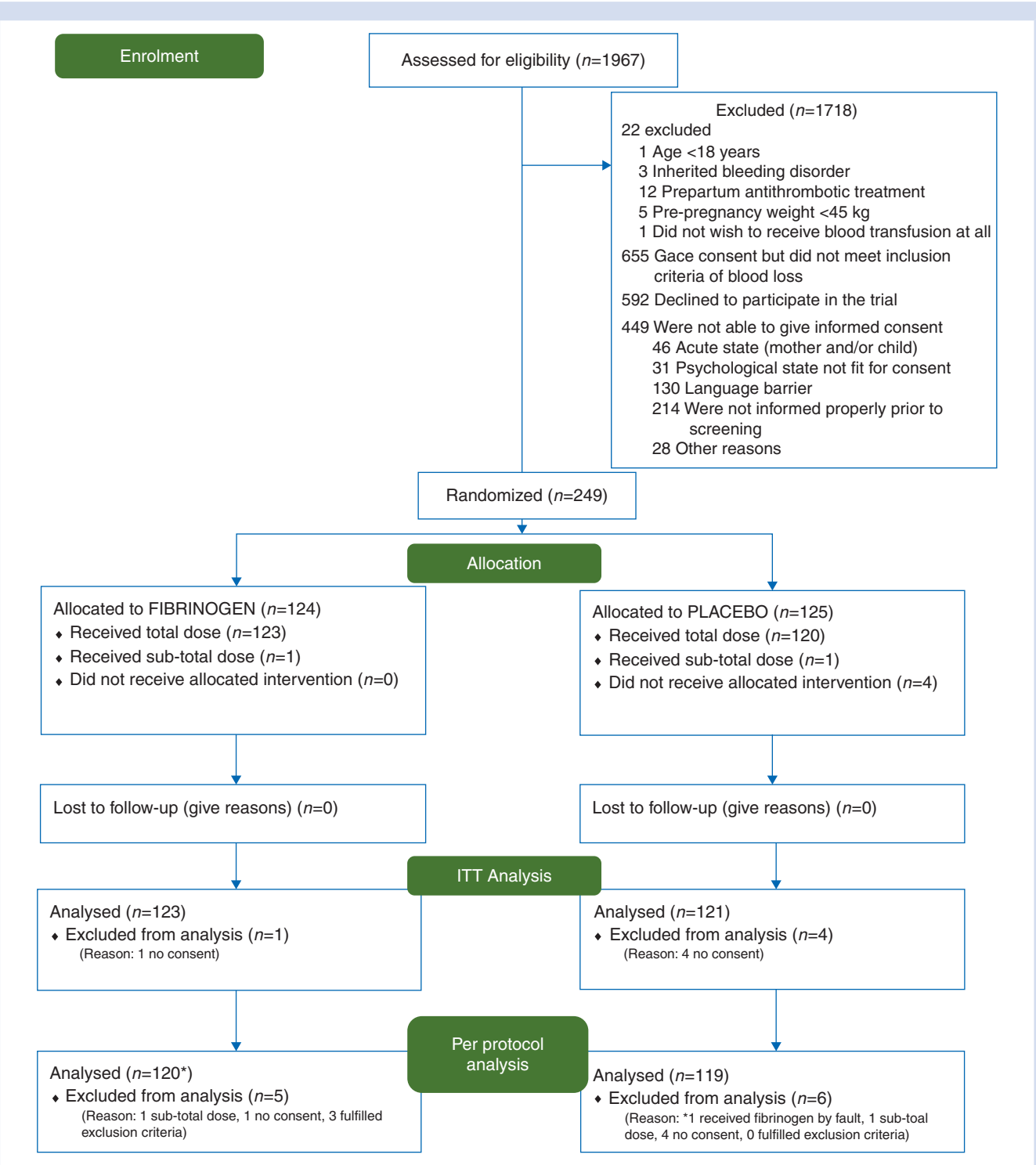

Fig 1 Consort flow diagram. ITT, intention to treat. *The subject who received fibrinogen despite allocation to placebo was included in the per-protocol analysis as part of the fibrinogen group (analysed as treated).

recommend transfusion with RBC if bleeding becomes uncontrollable (e.g. haemodynamic instability) or with significant anaemic symptoms provided haemoglobin (Hb) <72 g litre $^{-1}$ (Box 1).[14] Secondary outcomes were total blood loss, total amount of blood transfused, reoccurrence of bleeding, Hb <58 g litre^{-1}, RBC transfusion within 4 h, 24 h, and 7 days, and a composite outcome of 'severe PPH', defined by Charbit and colleagues[4] as a decrease in Hb of >40 g litre^{-1}, transfusion of ≥ 4 units of RBCs, haemostatic intervention (angiographic embolization, surgical arterial ligation, or hysterectomy), or maternal death.

Box 1 Summary of transfusion protocol.[11 13 14] RBC, red blood cell

Fluids

Initial infusion of 1–2 litres of crystalloids

If a plasma expander is needed, give human albumin 5%; avoid synthetic colloids

Blood transfusion

- Life-threatening haemorrhage (haemodynamic instability):
  Start balanced transfusion of RBC, fresh frozen plasma and platelets, aiming at a ratio of 1:1:1*
  Change strategy when haemodynamic stability is obtained
- Controllable bleeding (haemodynamic stability):
  Give RBC transfusion if haemoglobin is <72 g litre^{-1} (4.5 mM litre^{-1})
  Give fresh frozen plasma and platelets guided by thromboelastography (Kaolin TEG®: R time >11 min or angle <52 degrees and Maximum Amplitude <50 mm)

Tranexamic acid

Tranexamic acid 1 g i.v. should be considered early

*With Danish transfusion units, this corresponds to 5:5:2 of RBC: fresh frozen plasma:platelets

Statistical analyses

We calculated that 245 subjects were needed to show a reduction in relative risk of 33% with a power of 80%, a two-sided type I error of 0.05, and up to 15% dropouts. We estimated that fibrinogen would reduce the proportion of subjects receiving postpartum transfusion from 57 to 38%. The estimate of the proportion of subjects in need of RBC transfusion was based on approximately 1% (range 0.31–2.7%)[10] of parturients receiving RBC transfusion and 1.75% developing severe PPH (defined by a blood loss >1 litre).[15] Analyses of outcomes were performed before breakage of the randomization code. The intention-to-treat (ITT) population was used for analyses, including all randomized subjects with consent to participate regardless of exclusion criteria or having received the wrong dose. In the per-protocol analysis, we omitted those who met exclusion criteria or who did not receive the full dose. Estimates of treatment effect were compared for the pre-stated subgroups.[10] Unadjusted χ^2 tests for binary outcome measures and Student's *t*-test or Wilcoxon rank-sum test were used for continuous measures. Results are presented as the relative risk with a 95% confidence interval (CI), mean (SD), and *P*-value. Logistic regression was used to assess association with baseline variables, and we present adjusted analysis with odds ratio (OR), 95% CI, and *P*-value. Explanatory *post hoc* analyses using logistic regression were performed, including factors with baseline imbalance, statistical interaction, or suspected additional confounding effect. The stratification variable (centre) and baseline variables that were significant in the univariate analysis were included in the adjusted analyses. All analyses were performed with the use of R statistical software, version 3.0.2 (R Foundation for Statistical Computing, Vienna, Austria). A two-sided *P*-value of <0.05 was considered statistically significant. Changes in fibrinogen and Hb during the first 24 h after study drug infusion were compared between groups using longitudinal analysis (mixed-effect model).

Role of the funding source

This trial was funded by independent funds and without any financial or academic involvement from the manufacturer of fibrinogen concentrate. The funding source had no involvement. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit for publication.

Results

Study population

A total of 249 subjects were randomized, and 244 were available for final analysis on transfusion requirements up to 6 weeks; 123 in the fibrinogen group and 121 in the placebo group (Fig. 1). All 244 subjects had severe PPH, with a mean blood loss of 1.5 litre, and 10% with a systolic arterial pressure <100 mm Hg at inclusion. The distribution of subjects by inclusion criteria was as follows: exploration of the uterine cavity after vaginal delivery [$n=123$ (50%)], Caesarean section and blood loss >1 litre [$n=40$ (16%)], and decision on manual removal of a retained placenta with initial blood loss ≥ 0.5 litre [$n=81$ (33%); Table 1]. Three subjects (all assigned to the fibrinogen group) had exclusion criteria discovered following intervention; one with Factor Leiden V mutation, and two with antithrombotic treatment during pregnancy. Baseline characteristics are presented in Table 1. Baseline fibrinogen concentration was 4.5 g litre^{-1}, with 25% of values being subnormal for pregnancy (<3.7 g litre^{-1}),[3] but very few were <2 g litre^{-1} (2.2%). The single fibrinogen concentrate dose of 2 g of fibrinogen concentrate corresponded to a dose of 26 mg kg^{-1} and significantly increased plasma fibrinogen concentration. We obtained a mean difference of 0.40 g litre^{-1} between the groups 15 min after intervention (Fig. 2).

Intervention, placebo, and additional treatment

The mean fibrinogen dose administered to the intervention group was 26 (SD 4.2) mg kg^{-1} body weight at term. Tranexamic acid (median dose, 1 g; range, 1–2) was part of the standard treatment within 24 h in 92 subjects (38%), and five subjects (2%) received hydroxyethyl starch postintervention, with no significant difference between groups (Table 1). A similar amount of crystalloids (isotonic saline or Ringers acetate) were given postintervention, with a median infused volume of 1325 ml [inter-quartile range (IQR) 900–2000]. Longitudinal analysis (mixed-effect model) showed changes in fibrinogen during the first 4 h following study drug infusion. The fibrinogen group had a significantly higher fibrinogen concentration of 0.40 g litre^{-1} (95% CI, 0.15–0.65) at 15 min after study drug infusion compared with the placebo group (Fig. 2). Additional longitudinal analysis of Hb concentrations showed a significant decrease during the first 24 h compared with baseline, but with no difference between the fibrinogen group and the placebo group (details of additional treatment and fibrinogen concentration can be found in Fig. 2 and Supplementary data).

Table 1 Baseline characteristics of trial population (ITT). Data are presented as *n* (%) or mean (SD). HELLP, haemolysis, elevated liver enzymes, low platelet count; IQR, inter-quartile range; ITT, intention to treat; IU, international units; SD, standard deviation. *$P<0.05$, **$P<0.01$. †Predefined baseline characteristic according to protocol.[10] ‡'Start of bleeding' is defined as the time when the midwife recognizes blood loss exceeding the expected. Maternal weight at term is based on hospital charts and self-reported weight; weight of child is missing from calculations in the case of twins. ¶Cumulative group count and percentage

Characteristic		Fibrinogen (*n*=123)	Placebo (*n*=121)	
Inclusion criteria†				
Manual exploration of the uterus because of continuous bleeding after delivery of the placenta and blood loss ≥1 litre		67 55%	56 46%	
Manual removal of placenta and blood loss ≥0.5 litre		37 30%	44 36%	
Caesarean section and blood loss ≥1 litre		19 15%	21 17%	
Centre†	No. 1	24 20%	24 20%	
	No. 2	22 18%	22 18%	
	No. 3	27 22%	25 21%	
	No. 4	50 41%	50 41%	
Cause of haemorrhage (it is possible to have more than one cause of haemorrhage)				
Tone†		**68** **55%**	**54** **45%**	¶
Trauma†		**54** **44%**	**55** **46%**	¶
	Paravaginal haematoma	1 1%	1 1%	
	Laceration of cervix	11 9%	13 11%	
	Laceration of vagina	21 17%	21 17%	
	Laceration of perineum	33 27%	32 26%	
	Uterine rupture	0 0%	0 0%	
	Bleeding from uterotomy	6 5%	8 7%	
	Episiotomy	6 5%	1 1%	
Tissue†		**76** **62%**	**80** **66%**	¶
	Retained placental tissue	74 60%	78 65%	
	Placenta accreta or percreta	3 2%	4 3%	
	Placenta praevia	1 1%	1 1%	
Thrombin†		**10** **8%**	**13** **11%**	¶
	Diffuse Intravascular coagulopathy	0 0%	0 0%	
	Placental abruption	2 2%	2 2%	
	Pre-eclampsia	8 7%	12 10%	
	HELLP	0 0%	0 0%	
	Gestational thrombocytopenia	1 1%	0 0%	

Continued

Table 1 *Continued*

Characteristic		Fibrinogen (*n*=123)	Placebo (*n*=121)	
Time from delivery to start of bleeding‡ (min)†	Mean (SD)	35 (63)	28 (54)	
Obstetrical characteristics				
Parity	Multipara	58 47%	58 48%	
	Primipara	65 53%	63 52%	
No. of previous Caesarean sections	None	113 92%	107 88%	
	One	10 8%	14 12%	
Multiple gestation	Singleton	117 95%	118 98%	
	Twins	6 5%	3 3%	
Previous postpartum haemorrhage		14 11%	16 13%	
Antepartum bleeding		9 7%	14 11%	
Maternal weight at term (kg)†	Mean (SD)	79 (13)	82 (14)	*
BMI before pregnancy (kg m^{-2})	Mean (SD)	22.8 (3.3)	24.4 (4.5)	**
Weight of child (g)	Mean (SD)	3562 (574)	3625 (579)	
Gestational age at delivery (days)	Median [IQR]	282 [273;289]	284 [274;288]	
Type of anaesthesia†	General	27 22%	18 15%	
	Regional	96 78%	103 85%	
Status at inclusion				
Time from consent to randomization (h)	Median Range (min;max)	1 (0;70)	0 (0;67)	
Time from start of bleeding‡ to infusion of study drug (min)†	Median [IQR]	81 [59;130]	67 [46;115]	
Estimated blood loss at inclusion (ml)†	Mean (SD)	1493 (489)	1426 (463)	
Systolic arterial pressure (mm Hg)	Mean (SD)	122 (21)	124 (20)	
Systolic arterial pressure <100 mm Hg		14 11%	10 8%	
Diastolic arterial pressure (mm Hg)	Mean (SD)	73 (17)	72 (15)	
Heart rate (beats min^{-1})	Mean (SD)	98 (23)	99 (19)	
Haemoglobin at baseline (g $litre^{-1}$)†	Mean (SD)	105 (14)	103 (19)	
Incidence of initial hypofibrinogenaemia (baseline fibrinogen <2 g $litre^{-1}$ with Clauss method)†	Fibrinogen <2 g $litre^{-1}$	1 (1%)	4 (4%)	
	Fibrinogen >2 g $litre^{-1}$	119 99%	107 96%	
Initial fibrinogen concentration (g $litre^{-1}$)†	Mean (SD)	4.5 1.1	4.5 1.3	
Crystalloids given before inclusion (ml)†	Mean (SD)	1460 (796)	1298 (797)	

Continued

Table 1 *Continued*

Characteristic		Fibrinogen (*n*=123)	Placebo (*n*=121)	
Use of hydroxyethyl starch before inclusion†		1 1%	6 5%	
Transfusion of red blood cells before inclusion†		2 2%	0 0%	
Use of tranexamic acid before intervention†		40 33%	36 30%	
Tranexamic acid dose	Median [IQR]	1000 [1000;1000]	1000 [1000;1000]	
Oxytocin		117 95%	118 98%	
Oxytocin dose (IU)	Mean (SD)	15 (6)	15 (7)	
Misoprostol		91 74%	86 71%	
Misoprostol dose (mg)	Median [IQR]	0.4 [0.4;0.4]	0.4 [0.4;0.4]	
Methergine		32 26%	16 13%	*
Methergine dose (mg)	Median [IQR]	0.2 [0.2;0.2]	0.2 [0.2;0.2]	
Carboprost		27 22%	10 8%	**
Carboprost dose (mg)	Median [IQR]	0.2 [0.2;0.2]	0.2 [0.2;0.2]	

Outcome and adverse events

Red blood cell transfusion during the 6 week follow-up period postpartum was given to 25 (20.3%) of the fibrinogen group and 26 (21.5%) of the placebo group (relative risk, 0.95; 95% CI, 0.58–1.54; P=0.88; Table 2). Similar results were obtained in the per-protocol and the adjusted analysis (Supplementary data). No significant difference was found between transfusion incidences in the two groups at any time point recorded. No subjects received fresh frozen plasma or platelets. We found no difference in any secondary outcomes (Table 2) or registered adverse events (Table 3) between groups in either the ITT or the per-protocol analyses. We found no thromboembolic events. Trauma causing PPH, estimated blood loss, and Hb at baseline were significant risk factors for postpartum transfusion, but adjusted analyses did not change effect estimates of intervention (Supplementary data). Increased fibrinogen concentration following intervention was associated with a decreased risk of postpartum transfusion (OR, 0.65; 95% CI, 0.47–0.87; P=0.005), but this association was not significant (OR, 0.90; 95% CI, 0.60–1.34; P=0.61) when adjusted for estimated blood loss and Hb at baseline, dilution with crystalloids (at baseline and postintervention), hypovolaemia (systolic arterial pressure <100 mm Hg), centre, and cause of PPH (trauma or tissue). No change in effect estimates was found in the preplanned subgroups of subjects with vaginal or Caesarean deliveries. Too few subjects were available to assess a subgroup effect in patients with initial fibrinogen concentration <2 g litre^{-1}, and *post hoc* subgroup analysis in those with initial fibrinogen below the normal pregnancy concentration[3] (<3.7 g litre^{-1}) did not change effect estimates. No statistical interaction between initial fibrinogen concentration and effect of fibrinogen concentrate was identified.

Blinding

A total of 220 (94%) of the 235 anaesthetists who evaluated blinding following intervention had no idea of which treatment was provided, but nine (4%) had noticed a small amount of foam in the tubes, indicating fibrinogen concentrate. Besides the deliberate unblinding of one subject (placebo) who developed universal urticaria 2 days after the intervention, no subjects had discovered their allocation. Based on recovery or experience of adverse events, 98 (41%) patients felt they knew their allocation group. However, the same proportion believed they had received fibrinogen in both groups (Supplementary data). Sensitivity analysis of complete blinding did not affect estimates of treatment effect (Supplementary data).

Discussion

In this investigator-initiated, randomized, double-blinded, multicentre trial, we found no reduction in RBC transfusion during a 6 week follow-up period postpartum following administration of 2 g of fibrinogen concentrate.

The FIB-PPH trial is the largest randomized controlled trial investigating fibrinogen concentrate.[8] It is investigator initiated, the first trial in obstetric patients, and the only trial

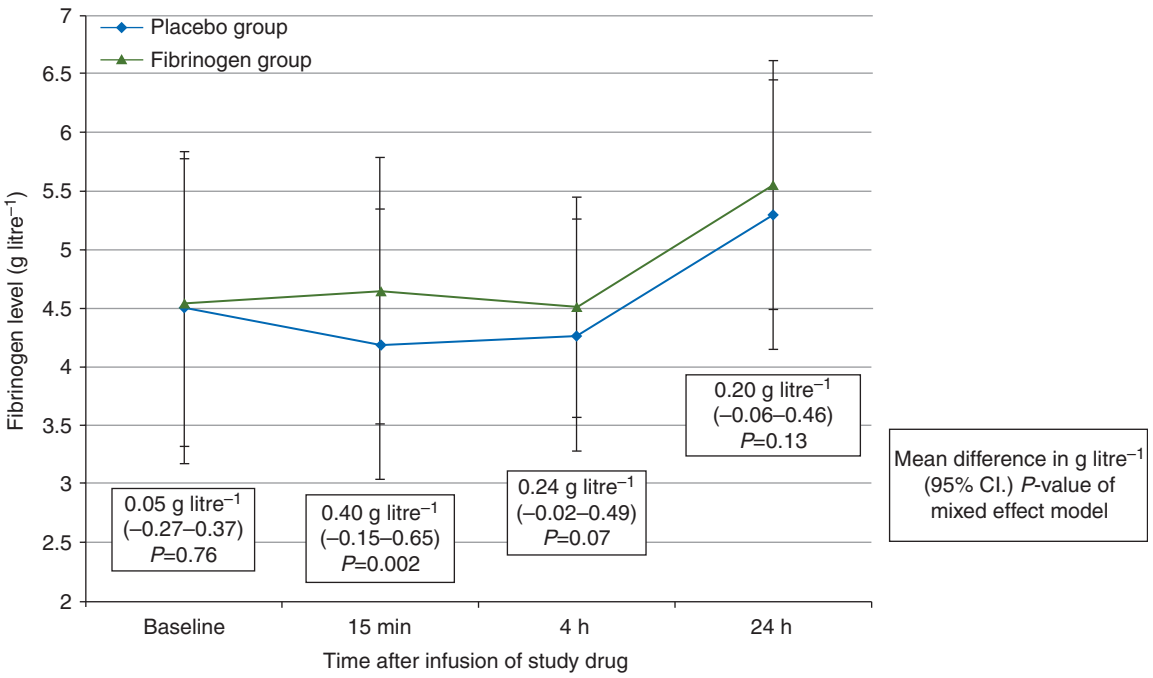

Fig 2 Mean fibrinogen concentrations in placebo and fibrinogen groups from baseline to 24 h after study drug administration, with whiskers indicating standard deviation. Mean difference of the fibrinogen concentration between the fibrinogen and placebo group is given below at each time point from baseline to 24 h after the study drug administration, with 95% confidence interval (CI) given in parenthesis and *P*-value.

Table 2 Primary and secondary outcomes, intention to treat. RBC, red blood cell. Data are presented as the median [IQR] or *n* (%). *One hundred and forty-eight values are missing (61%). †Mean difference with 95% confidence interval (CI; Student's *t*-test). ‡Wilcox rank sum test

Outcome	Fibrinogen (*n*=123)	Placebo (*n*=121)	Relative risk (95% CI)	*P*-value
Primary outcome				
Need for RBC transfusion (during the 6 week period postpartum)	25 (20.3%)	26 (21.5%)	0.95 (0.58–1.54)	0.88
Secondary outcomes				
Estimated blood loss after study drug (ml)	1700 [1500–2000]	1700 [1400–2000]	66 [−78; 210]†	0.37
Need for RBC transfusion (up to 4 h after study drug)	4 (3.3%)	10 (8.3%)	0.39 (0.13–1.22)	0.11
Need for RBC transfusion (up to 24 h after study drug)	14 (11.4%)	19 (15.7%)	0.72 (0.38–1.38)	0.35
Need for RBC transfusion (up to 7 days after study drug)	25 (20.3%)	26 (21.5%)	0.95 (0.58–1.54)	0.88
Total amount of blood transfused	0 [0,0]	0 [0,0]	‡	0.83
Range [min, max]	[0,7]	[0,4]		
Severe PPH*	20 (40.0%)	24 (52.2%)	0.77 (0.49–1.19)	0.31
Death	0 (0.0%)	0 (0.0%)	–	
Haemostatic intervention	0 (0.0%)	0 (0.0%)	–	
Transfusion of ≥4 units of RBCs	8 (6.5%)	3 (2.5%)	2.62 (0.71–9.65)	0.22
Decrease in haemoglobin >40 g litre^{-1}*	20 (40.0%)	24 (52.2%)	0.77 (0.49–1.19)	0.31
Rebleeding	2 (1.6%)	2 (1.7%)	0.98 (0.14–6.87)	1.00
Lowest haemoglobin <58 g litre^{-1}	1 (0.8%)	5 (4.1%)	0.20 (0.02–1.66)	0.12

where subjects were randomized in an emergency setting. Only two of six previous randomized controlled trials investigating fibrinogen concentrate blinded subjects and clinicians,[8] but in this trial we successfully blinded most clinicians and all the subjects. The pragmatic multicentre trial set-up and our few exclusion criteria strengthen the external validity, with the randomized design reducing selection and performance bias. However, our results are limited by the wide confidence intervals on the primary result caused by the lower than expected proportion of included subjects in need of RBC transfusion.[10] The primary reason for this is probably the inability to include subjects with massive and rapid bleeding because of

Table 3 Adverse events, intention to treat. Data are presented as *n* (%). *One hundred and forty-eight values are missing (61%)

	Fibrinogen (*n*=123)	Placebo (*n*=121)	Relative risk (95% CI)	*P*-value
Adverse events				
Up to 24 h				
Dizziness	27 (22.0%)	33 (27.3%)	0.80 (0.52–1.25)	0.37
Shivering	7 (5.7%)	6 (5.0%)	1.15 (0.40–3.32)	1.00
Fever (>38.0°C)	12 (9.8%)	8 (6.6%)	1.48 (0.63–3.48)	0.49
Abdominal pain	10 (8.1%)	3 (2.5%)	3.28 (0.93–1.62)	0.08
Headache	12 (9.8%)	10 (8.3%)	1.18 (0.53–2.63)	0.82
Nausea or vomiting	6 (4.9%)	6 (5.0%)	0.98 (0.33–2.97)	1.00
Fainting	5 (4.1%)	2 (1.7%)	2.46 (0.49–12.43)	0.45
Palpitations	3 (2.4%)	5 (4.1%)	0.59 (0.14–2.42)	0.50
Allergic reaction	0 (0.0%)	1 (0.8%)	–	
Urticaria	0 (0.0%)	1 (0.8%)	–	
Itching	2 (1.6%)	0 (0.0%)	–	
Shortness of breath	1 (0.8%)	3 (2.5%)	0.33 (0.04–3.11)	0.37
Facial oedema	1 (0.8%)	2 (1.7%)	0.49 (0.05–5.35)	0.62
Six week follow-up				
Thromboembolic complications	0 (0.0%)	0 (0.0%)	–	
Readmission with the need for re-evacuation of uterine cavity	1 (0.8%)	2 (1.7%)	0.49 (0.05–5.35)	0.62

the need for informed written consent obtained in the emergency setting. As a result of our early pre-emptive treatment regimen, inclusion was not restricted to patients with hypofibrinogenaemia, and fibrinogen dose was not adjusted to body weight or haemostatic measures. Included patients should be able to give informed consent. The median time from screening to inclusion was <1 h. This reflects that most subjects delivered vaginally, not by planned Caesarean section, and therefore consent was obtained when epidural anaesthesia was provided or when PPH was diagnosed. The use of antepartum written information and obtaining final consent was in accordance with the recommendations of Royal College of Obstetricians and Gynaecologists.[16] In 46 screened patients, it was impossible to obtain consent. Not being able to include cases with severe and rapid PPH is probably reflected in the low number of subjects with initial hypofibrinogenaemia and a lower incidence of RBC transfusion than expected. The incidence of venous thromboembolic events in our population is estimated to be 0.7–2 per 1000 pregnancies.[17] The sample of 244 subjects is insufficient to assess the risk of thrombosis associated with fibrinogen concentrate, but no symptomatic thromboembolic events were seen at the 6 week follow-up.

A Cochrane review showed that fibrinogen concentrate in bleeding patients was associated with a 53% reduction in RBC transfusion,[8] but it included only 208 patients, mainly undergoing cardiac surgery. The fibrinogen concentration was <2 g litre^{-1} at inclusion, except for one study investigating preoperative prophylactic treatment of patients with a mean preoperative fibrinogen of 2.9 g litre^{-1}.[18] Few of our subjects received colloids, and given that fibrinogen concentrate can alleviate the haemostatic impairment of synthetic colloids (e.g. hydroxyethyl starch),[19] this could explain the lack of efficacy in our trial compared with trials in the Cochrane review. Tranexamic acid reduces fibrinolysis and protects endogenous fibrinogen. Tranexamic acid was given to 31% of subjects before intervention and few had a fibrinogen <2 g litre^{-1} at inclusion, and thus they might have had a lesser degree of initial coagulopathy compared with patients after cardiopulmonary bypass.[20] A total of 38% of subjects received tranexamic acid within 24 h, and even if our adjusted analysis did not detect a direct influence on blood transfusion it might be that co-administration of tranexamic acid would have given another result.[9] As other co-interventions can dilute fibrinogen and thus affect the validity of our findings, we have provided detailed description of co-interventions and aimed to reduce their impact on our findings. The change in fibrinogen before and after study drug administration is not only a result of the study treatment, but mainly because of ongoing dilution by fluids (mainly crystalloids) and consumption (e.g. fibrinogen concentration decreased by 0.3 g litre^{-1} in the placebo group). Fibrinogen concentration was 0.4 g litre^{-1} higher in the fibrinogen group following administration of study drug compared with placebo. The fibrinogen increment of 0.1 g litre^{-1} obtained in the fibrinogen group shows that 2 g of fibrinogen concentrate was enough to avoid a decrease, and even to restore and increase fibrinogen concentration. In the study by Charbit and colleagues,[4] 10% of patients with PPH after manual exploration of the uterine cavity requiring i.v. prostaglandin administration had an initial fibrinogen concentration <2 g litre^{-1}. Unfortunately, no data were presented on blood loss or resuscitation with crystalloids or colloids. In our study, fewer subjects had an initial fibrinogen concentration <2 g litre^{-1}, fewer received fresh frozen plasma and were in need of haemostatic intervention, but an equal proportion

presented with hypovolaemia and a higher rate of RBC transfusion within 24 h. Charbit and colleagues[4] reported a longer delay from inclusion to baseline blood sampling in those patients who developed the most severe course of bleeding;[4] this delay might be associated with increased dilution, loss of blood, and loss of fibrinogen such that the initial concentration of fibrinogen may be a surrogate marker of blood loss.[21] This corresponds to our finding that the risk of postpartum transfusion was lower in those with increased fibrinogen concentration following intervention, but this effect disappeared after adequate adjustments for dilution, blood loss, tranexamic acid use, and the cause of PPH.

Fibrinogen concentrate is increasingly used in acquired hypofibrinogenaemia associated with bleeding and as pre-emptive treatment in PPH.[22] Some even recommend 2–4 g of fibrinogen concentrate in the case of 1.5 litre bleeding.[23] However, this represents off-label use in most countries.[24] Several observational studies have reported on the use of fibrinogen concentrate in obstetrical haemorrhage;[25–30] in patients with low fibrinogen (1–1.5 g litre^{-1}) and co-administration of multiple transfusions, including fresh frozen plasma. These studies are based on retrospective chart evaluation, without appropriate control groups, and are associated with a high risk of selection bias, especially the risk of confounding by indication. In addition, most trials investigating fibrinogen concentrate have been sponsored by the manufacturer of fibrinogen concentrate.[8] Based on our findings, further pre-emptive use of fibrinogen concentrate in PPH[22] is not justified. Future studies on PPH should investigate the impact of goal-directed fibrinogen substitution in patients with hypofibrinogenaemia and apply a method of consent that allows for inclusion of patients with rapid and massive bleeding.

In conclusion, we found no evidence for the use of pre-emptive treatment with fibrinogen concentrate for severe postpartum haemorrhage in patients with normofibrinogenaemia.

Supplementary material

Supplementary data are available at *British Journal of Anaesthesia* online.

Authors' contributions

A.J.W., H.M.E., A.A., J. Stensballe, J.L.-R., and A.M.M. developed the concept and trial design, and served as steering and writing committee. A.J.W. and H.M.E. served as trial managers, collected and validated data, and secured funding. A.J.W. designed data collection tools and wrote the statistical analysis plan, cleaned and analysed the data, and drafted and revised the paper; she is guarantor. Analyses were recalculated by independent statistician and data manager Tobias Wirenfeldt Klausen, Clinical research Unit, Department of hematology, Herlev Hospital, University of Copenhagen, Denmark. C.A., K.E., G.H., E.L.S., H.F.S., A.U.M., L.F., J. Svare, A.T., L.M.P., J.L., M.G.M., and B.B. made substantial contributions to the implementation of the trial at each centre, the enrolment of patients, the solving of logistics, and the overall concept of the trial. All authors were involved in drafting of the manuscript and revising it critically. All have approved the final version. All authors had full access to data, including statistical reports and tables, and can take responsibility for the integrity of the data and the accuracy of the data analysis.

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Acknowledgements

We would like to thank all participating women and their relatives together with the staff at all sites, including midwives, obstetricians, anaesthetist nurses and doctors, personnel at the Blood Bank, and technical personnel at departments of anaesthesia, Herlev Hospital (Belinda Ierst, Ulla Jensen, Helle Christensen and Bente Lauridsen) together with Anne Marie Münster, Department of Biochemistry at Aalborg Hospital. Thanks to our blood samples response team of medical students and the monitoring unit of Good Clinical Practice at Bispebjerg Hospital, especially Lotte Lauersen. Thanks also to Professor Lars Rasmussen, Department of Anaesthesia, Centre of Head and Orthopedics, Rigshospitalet, Denmark for critical revision of the manuscript. We would also like to thank the independent funding body, Haemonetics, and the Heads of all departments for making it possible to conduct this trial.

Declaration of interest

None declared.

Funding

The Danish Council for Independent Research | Medical sciences; Grant ref. no. 10-084494; Region Hovedstadens Sundhedsvidenskabelige Forskningsfond; Forskningsrådet Herlev Hospital; Laerdals Acute Foundation; Aase og Ejnar Danielsens Fond, Fonden af 17-12-1981; Fonden til lægevidenskabens fremme/ved A.P. Møller og Hustru Chastine Mc-Kinney Møllers Fond til almene Formaal; Hans and Nora Buchards fond. Thromboelastography kits for assays were provided by Haemonetics Corp., Braintree, MA, USA.

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Appendix

The FIB-PPH trial group

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Handling editor: H. C. Hemmings

Massive postpartum transfusion: a multidisciplinary observational study

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Abstract

Objective

To describe the severe and rare cases of postpartum haemorrhage (PPH) requiring massive transfusion including the surgical procedures performed, causes, and complications in these women. Furthermore to describe the subgroup of women requiring peripartum hysterectomy during massive transfusion and compare the use of Red Blood Cell (RBC), Fresh Frozen Plasma (FFP) and Platelet (PLT) transfusions with women not requiring hysterectomy during massive transfusion.

Design

Cohort study.

Setting

Danish hospitals with maternity units.

Population

All pregnant women receiving ≥10 units of RBC in a 24 hour period up to 6 weeks postpartum from January 2001 to December 2009.

Methods

Review of all patient charts identified by combining The Danish Transfusion Database with The Danish Medical Birth Registry.

Results

A total of 245 women received massive postpartum transfusion of which 128 (52%) required a hysterectomy. The main causes of massive postpartum transfusion were atony (38%), abnormal invasive placenta (25%), unintended extension of the uterine incision (24%) and placenta praevia (24%), with 34% developing coagulopathy at some point of time. Complications included 2 deaths, 6 cardiac arrests, 3 with renal failure requiring dialysis, and 100 in need of ventilator support after surgery. Women requiring a hysterectomy received a higher FFP:RBC ratio (p=0.010) and received more units of RBCs before their first PLT transfusion (p=0.023) than women not requiring a hysterectomy. Hysterectomy led to haemostasis in 90 women (70%).

Conclusions

Women requiring massive postpartum transfusion had a high incidence of severe morbidity including hysterectomy. Hysterectomy is generally considered last treatment option guaranteed to secure haemostasis, but only 70% of the hysterectomies with massive postpartum transfusion resulted in haemostasis. Women treated with a hysterectomy had increased blood loss and increased transfusion requirements at the time of haemostasis compared to women not requiring hysterectomy.

Introduction

Postpartum haemorrhage (PPH) remains one of the leading causes of maternal morbidity and mortality worldwide.[1–3] PPH can in severe cases with excessive bleeding lead to blood transfusions, multiple operations, organ failure, shock, disseminated intravascular coagulation (DIC), and maternal death.[4–6] The need for massive transfusion defined as the need of ≥10 RBC units in patients with severe or uncontrollable haemorrhage is only seen in approximately 0.6/1,000 deliveries.[6,7] Under these circumstances the pathophysiological changes due to replacement of total blood volume are pronounced and usually associated with the development of substantial coagulopathy.[7,8] This severe impairment of haemostasis can be dilutional due to transfusion of red blood cells (RBC), colloids and crystalloids, and consumptive due to activation of the coagulation cascade as a result of hypoperfusion and tissue damage.[8,9]

Haemostatic resuscitation with early transfusion of fresh frozen plasma (FFP) and platelets (PLT) has been introduced to reduce the development of coagulopathy in patients with massive haemorrhage. The concept of haemostatic resuscitation is to use a fixed ratio of blood products and to avoid use of colloids and crystalloids that can further aggravate coagulopathy by dilution.[9–11] Recent studies in trauma patients receiving massive transfusions have shown a reduced risk of haemorrhagic death and increased early survival when a fixed ratio of 1:1:1 of PLTs, FFP and RBCs is used.[12,13] Outside trauma, a fixed ratio of 1:1:1 is associated with higher survival in ruptured aortic

aneurisms and in a mixed group of massive haemorrhage including PPH.[14,15] Specific evidence for PPH is limited, but early and high dose use of plasma has been associated with a reduced risk of haemostatic interventions (interventional radiology and hysterectomy).[16] In contrast to trauma studies the low incidence of mortality in women with PPH renders mortality a difficult outcome. Instead hysterectomy due to bleeding could be considered a relevant outcome as a sign of morbidity and the last resort in treating women with massive postpartum transfusion.

In the present study we used two national databases to identify women that received ≥10 RBC units in a 24 hour period postpartum, validating the data through detailed review of the patient charts. The objective was to describe these severe and rare cases of PPH with massive transfusion including the surgical procedures leading to bleeding control and the causes and complications associated with severe bleeding. Furthermore, we wanted to describe the subgroup of women requiring peripartum hysterectomy during massive transfusion and compare the use of RBC, FFP and PLT transfusions with women not requiring hysterectomy during massive transfusion.

Methods

Study population

This large cohort study was established by using Danish civil registration numbers to combine data from two national databases: The Danish Medical Birth Registry and The Danish Transfusion Database. We acquired permission from the Danish Data Protection Agency (Journal nr.2007-58-0015). No permission was required from the Danish Ethics Committee according to Danish legislation. The Danish Medical Birth Registry receives information regarding all births in Denmark and contains information regarding maternal demographics, parity, pregnancy, labour and delivery.[17] The Danish Transfusion Database collects information from regional blood banks and from The Danish National Patient Registry with regard to allogenic transfusions including data on the recipient, serial number of the blood components and time of delivery of blood products.[18] The database has systematically collected information since 1997, but was not complete for all hospitals in Denmark until 2005. Deliveries in hospitals not included in The Danish Transfusion Database were therefore excluded from our analysis up until 2005. We included all deliveries (singleton and multiple pregnancies) from 2001 to 2009 in the need of ≥10 RBC units within a 24-hour period up to 6 weeks postpartum and merged data from the two databases for all included patients. Subsequently, we manually collected and reviewed all available patient charts from all hospitals in Denmark for data validation and further data extraction. We excluded women receiving blood transfusions due to non-obstetric causes such as heart surgery, amputation, oncology surgery or sepsis postpartum (9 women), and women without accessible patient charts (4 women). Due to this study being planned in 2010, we were only able to extract data up to 2009.

Variables

Patient charts were used as the main source of data extraction. The first author HME extracted all data and conferred with co-authors in cases of uncertainty. Variables extracted included: Maternal age, parity, gestational age, site of birth, previous caesarean section, previous PPH, fertility treatment, hypertensive disorders, multiple gestation, placenta praevia, abnormal invasive placenta (AIP), medical induction of labour, augmentation of labour, antepartum haemorrhage, placental abruption, mode of delivery, reason for non-vaginal delivery, neonatal outcome, causes of onset of PPH, subsequent causes of PPH emerging after onset, time of birth, PPH, and haemostasis, blood loss and transfusions of RBCs, FFP, PLTs, colloids, crystalloids, fibrinogen concentrate, and recombinant factor VIIa before and after each surgery, uterotonics, surgical procedures performed and their rate of surgical control of the haemorrhage. Other variables included were: time in an Intensive Care Unit, intubation time, dialysis, perioperative assistance from other specialties, cardiac arrest, and death. It was not possible to verify BMI and birthweight from the patient charts, but they were included in the analysis.

Hysterectomy was defined as our primary outcome. Causes were divided into primary and secondary causes of onset and primary and secondary subsequent causes of haemorrhage. Surgical control of the bleeding was defined by haemostasis without the need for further major intervention. Time of haemostasis was defined when active bleeding was no longer suspected. Coagulopathy was defined either by direct diagnosis in the patient chart, or by a description of oozing from mucous membranes, puncture sites or raw surfaces. FFP:RBC ratio was defined as number of FFP transfusions divided by number of RBC transfusions. In addition we registered number of RBC transfusions before first FFP and number of RBC and FFP transfusions before first PLT transfusion Abnormally invasive placenta (AIP) was defined either by a direct diagnosis in the patient chart or by description of the placenta being torn apart upon manual removal. Life threatening haemorrhage was defined if either “uncontrollable haemorrhage” or “haemorrhage threatening the woman’s life” was noted in the patient charts. First surgery was defined as surgery in an operating theatre and included caesarean section or first surgery after vaginal delivery and could include multiple procedures. Caesarean section was included as first surgery because many haemostatic procedures were performed during caesarean section. End of surgery was defined as a closed laparotomy or conversion from a vaginal approach to a laparotomy. Haemostasis surgery was defined as the surgery where haemostasis was achieved. Major pregnancy complication was defined as: AIP, placenta praevia, HELLP syndrome (Haemolysis, Elevated Liver enzymes, Low Platelet count), uterine rupture or placental abruption. End organ dysfunction was defined as cardiac arrest, ventilator support ≥48 h after surgery, or renal failure (dialysis). The exact timing of each transfusion during surgery was not possible to decipher from the patient charts, as many transfusions were given simultaneously or given without exact documentation of

timing. Number of transfusions was instead estimated before, during, and after the surgery that led to haemostasis and also at the end of surgery where a hysterectomy was performed. It was not possible to give a solid estimate of transfusions prior to the actual hysterectomy, as the exact timing of this procedure was not documented, but could be part of several procedures performed back to back. In Denmark FFP needs to be thawed before use at all hospitals but one, and PLT transfusions consist of pooled buffy coat platelets from 4 donors.[19] By using data from patient charts and databases, where data are collected at time of event we were able to limit performance bias. Furthermore by including all hospitals in Denmark for the majority of the study period we were able to limit selection bias.

Statistical analysis

All analyses were performed using the statistical software SPSS 22.0 (SPSS inc, Chicago, IL, USA). Data are presented as proportions and percent (%), means and standard deviations (SD), or medians and interquartile ranges (IQR) for normally and non-normally distributed data. Univariate logistic regression analysis was used determine differences between women treated with and without hysterectomy during massive transfusion. A two-tailed p-value of less that 0.05 was considered statistically significant.

Results

Maternal, delivery characteristics

Massive PPH requiring massive transfusion complicated 245 out of 486,431 deliveries equivalent to 0.5 in 1,000 births from 2001 to 2009 in 29 different hospitals in Denmark (Figure 1). The majority of women were under 35 years old (67%), had a BMI of less than 25 (68%), were multiparous (63%), had term pregnancies (67%), and delivered live healthy babies (93%) by caesarean section (67%). A total of 112 women (46%) had one of the following major pregnancy complications: AIP, placenta praevia, HELLP syndrome, uterine rupture or placental abruption. However, only 6 of the 64 cases of AIP (9%) were identified before labour even though 46 (72%) had a prior caesarean section and/or placenta praevia (Table 1).

Characteristics of haemorrhage, transfusions and complications

Onset of haemorrhage was evenly distributed between night and day. The median time from birth to onset was 0:00 (hours:minutes) with 30% debuting before childbirth. The mean blood loss before first surgery after vaginal delivery was 1718ml (SD ±955ml). Few women received transfusions before first surgery (6%), but 17% had received at least 1,000ml of crystalloids. The median number of RBC transfusions before transfusion of FFP and PLT was 5 [IQR 4-8] and 8 [5-12]. At the end of the surgery that led to haemostasis involving multiple procedures, the women had received a median of 13 [10-18] RBCs, 6 [4-10] FFP and 1 [0-3] PLTs. The maximum number of RBC transfusions in one single woman was 108 with 57 women (24%) receiving more than 20 RBC transfusions each. Median blood loss was 8,000 ml (IQR 6,500-10,375) with 25% bleeding more than 10,000ml and one woman's blood loss estimated at 53,000 ml. The mean FFP:RBC ratio in the whole cohort was 0.48 (±0.23) at the time of haemostasis, with a significant rise from 0.41 (±0.28) in 2001 to 0.62 (±0.29) in 2009, p=0.005. The median total amount of crystalloids and colloids given was 7,000 [IQR 5,000-8,500] and 2,000 [IQR 1,500-2,500] respectively. A total of 21% received fibrinogen concentrate and 19% received recombinant factor VIIa (Table 2 and S1).

A total of 170 women (70%) were admitted to the Intensive Care Unit for at least 24 hours. End organ dysfunction developed in 109 women (45%), 2 with cardiac arrest that died, 6 with ventilator support following cardiac arrest, 2 with renal failure and ventilator support, 1 with renal failure alone, and 98 with ventilator support alone.

Figure 1: Flow chart of included women in the cohort

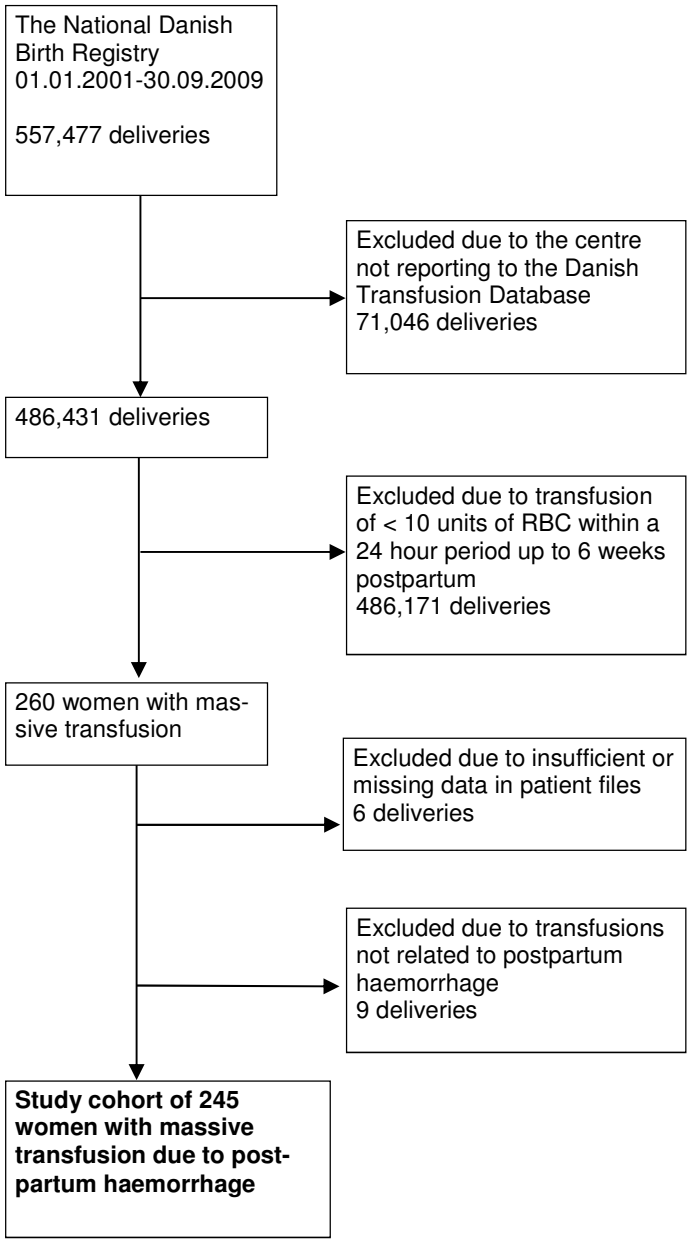


Table 1: Maternal and delivery characteristics of women with massive postpartum transfusion (depicted as proportions and percent (%)).

Maternal and delivery characteristics	n (%)
Age at partum	
<35	163 (66.5)
≥35	82 (33.5)
BMI (missing n=73)	
<25	117 (68.0)
25-35	48 (27.9)
>35	7 (4.1)
Gestational age (missing n=12)	
<37	54 (23.2)
37-42	157 (67.4)
>42	22 (9.4)
Parity (n=245)	
0	91 (37.1)
1-3	130 (53.1)
≥4	24 (9.8)
Previous caesarean section	74 (30.2)
Preeclampsia	21 (8.6)
Twins	14 (5.7)
HELLP syndrome	11 (4.5)
Praevia	43 (17.5)
Abnormal invasive placenta (identified before labour)	6 (2.4)
Abnormal invasive placenta (total)	64 (26.1)
Placental abruption	21 (8.6)
Uterine rupture	16 (6.5)
Major pregnancy complication	112 (45.7)
Mode of delivery	
Spontaneous vaginal	63 (25.7)
Instrumental	19 (7.8)
Planned caesarean section	38 (15.5)
Emergency caesarean section	125 (51.0)
Neonatal outcome (missing n=1)	
Live birth	228 (93.4)
Still birth	10 (4.1)
Died at or shortly after delivery	6 (2.5)
Birthweight > 4,000g	44 (18.0)

BMI = Body Mass Index, HELLP syndrome= Haemolysis, Elevated Liver enzymes, Low Platelet count, Major pregnancy complication: Abnormal invasive placenta, placenta praevia, HELLP syndrome, uterine rupture or placental abruption.

Other severe complications due to surgery included four injuries to the liver or spleen and one ligation of the external iliac artery by mistake. Median hospitalization was 9 days (IQR 6-14) (Table 2).

Procedures and their success rate

Figure 2 illustrates the many different procedures and combinations of procedures were performed in order to gain haemostasis. A median of 2.7 (IQR 2.0-3.7) procedures were performed during each surgery with a range from 1-10. The most widely used procedures were hysterectomy (n=128; 52%), suturing of genital tract lacerations (n=95; 39%), intrauterine palpation (n=85; 35%), suturing of the uterotomy (n=76; 31%), B-lynch suture (n=71; p=29%), vaginal tamponade (n=70; 29%), and bimanual compression of the uterus (n=65; 27%). The procedures performed that gained haemostasis most frequently (often in combination) were splenectomy (n=4; 100%), hysterectomy (n=90; 70%), embolization (n=2; 67%), suturing of the vaginal vault (n=17; 65%), ligation of the internal iliac artery or vein (n=5; 63%), and intraabdominal tamponade. Some of these procedures were however performed quite rarely.

Causes of haemorrhage

Figure 3 illustrates the wide variety of primary causes of onset of haemorrhage, and subsequent causes after onset of haemorrhage. The majority were due to atony (n=93; 38%), AIP (n=62; 25%), genital tract lacerations (n=60; 24%), unintended extension of the uterine incision (n=59; 24%), retained tissue (n=42; 17%), placenta praevia (n=33; 13%), and placental abruption (n=21; 9%) with many women having multiple causes. Out of the 146 women that developed subsequent causes after onset of haemorrhage, the majority developed atony (n=77; 53%), coagulopathy (n=31; 21%) or haematomas (n=17; 12%).

The different categories of primary cause of start of haemorrhage needed various different procedures performed to obtain haemostasis. Hysterectomy played the largest role in gaining haemostasis in AIP and/or placenta praevia (55%), placental abruption (43%) and retained tissue/placenta cases (41%) compared to all other procedures. In women where the primary cause was atony, haemostasis was obtained by either hysterectomy (30%), B-lynch suture (23%) or a procedure performed after hysterectomy (21%). Haemostasis for genital tract lacerations was obtained by hysterectomy (25%), suturing of genital tract lacerations (19%) or a procedure performed after hysterectomy (19%), while haemostasis for unintended extension of the uterine incision was obtained by hysterectomy (30%), B-lynch suture (16%) or a procedure performed after hysterectomy (12%) (Data not shown).

Table 2: Characteristics of PPH, transfusions and complications in women with massive postpartum transfusion (proportions and percent (%) are depicted for categorical data, means and standard deviations (SD) for normally distributed data and medians and interquartile ranges (IQR) for non-normally distributed data).

Characteristics	n (%), mean ± SD, median [IQR]
Characteristics of PPH	
Time of day haemorrhaging started	
08.00-18.00	117 (48.1)
18.00-08.00	126 (51.9)
Haemorrhage before birth (missing n=3)	72 (29.8)
Time from birth to start of haemorrhage (hours:minutes) (missing n=3)	0:00 [-0:07-0:08]
Blood loss before first surgery	
Vaginal delivery (ml)	1718 ± 955
Caesarean section (ml)	229 ± 541
Characteristics of transfusions	
Number of RBCs before first FFP	5 [4-8]
Number of RBCs before first PLT	8 [5-12]
Preoperative crystalloids	
≤ 1,000ml	202 (82.8)
>1,000ml	42 (17.2)
Number of RBCs at end of haemostasis surgery	13 [10-18]
Number of FFP at end of haemostasis surgery	6 [4-10]
Number of PLT at end of haemostasis surgery	1 [0-3]
FFP:RBC ratio at end of haemostasis surgery (missing n=7)	0.48±0.23
Complications	
Hospitalization time (days:hours:minutes) (missing n=10)	09:01:30 [6:14:00-14:04:40]
Total blood loss (ml) (missing n=17)	8,000 [6,500-10,375]
>10,000	57 (25.0)
End organ dysfunction	109 (44.5)
Post-operative intubation ≥ 2 hours (missing n=6)	106 (43.3)
Intensive care unit stay (missing n=3)	
<1 day	72 (29.8)
≥ 1 day	170 (70.2)
Renal dialysis treatment	3 (1.2)
Cardiac arrest	8 (3.3)
Death	2 (0.8)

PPH = postpartum haemorrhage, RBC = Red Blood Cell, FFP = Fresh Frozen Plasma, PLT = Platelet

Haemostasis surgery was defined as the surgery where haemostasis was achieved.

Figure 2: Procedures performed and their rate of haemostasis in women with massive postpartum transfusion.

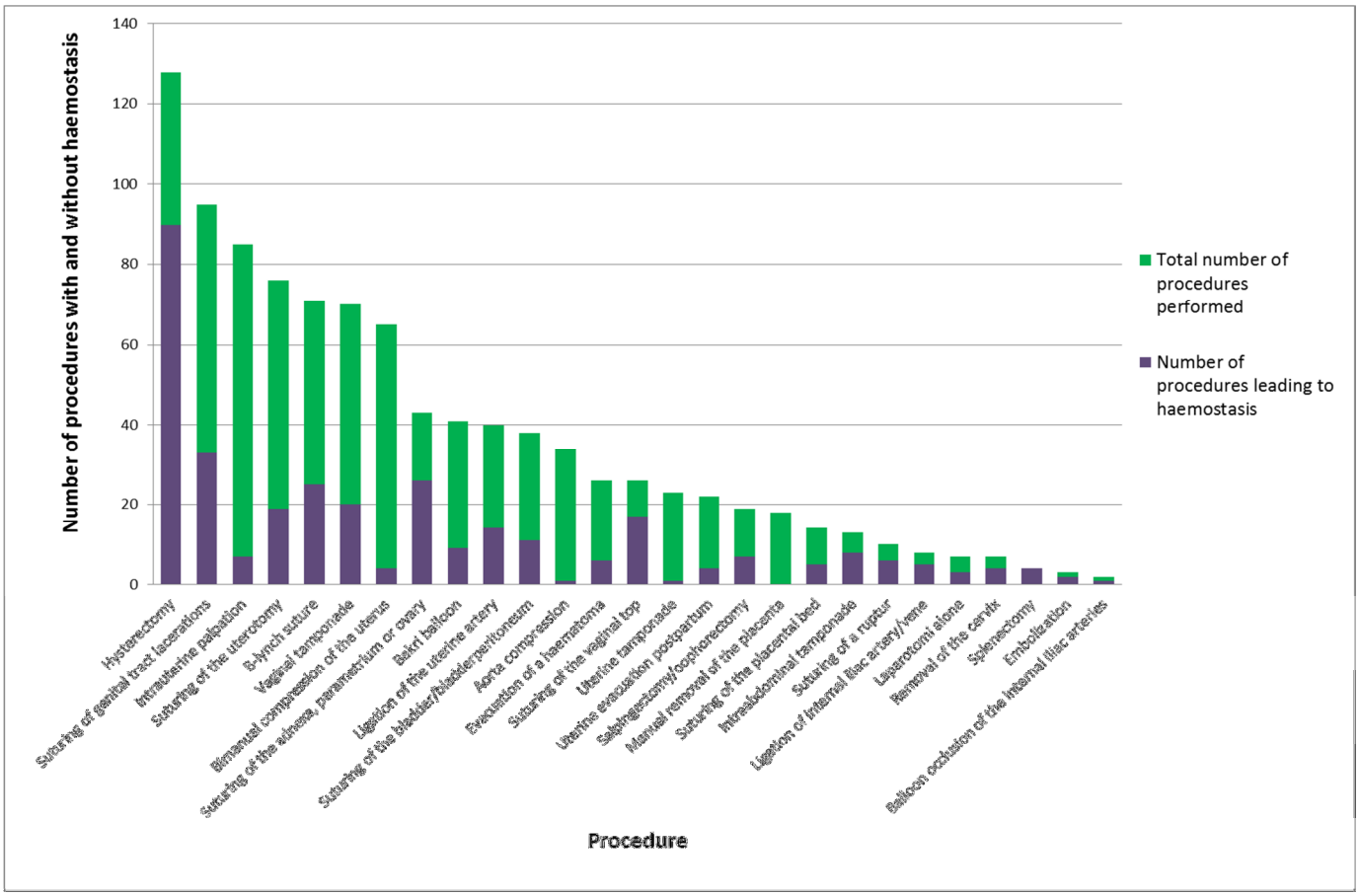
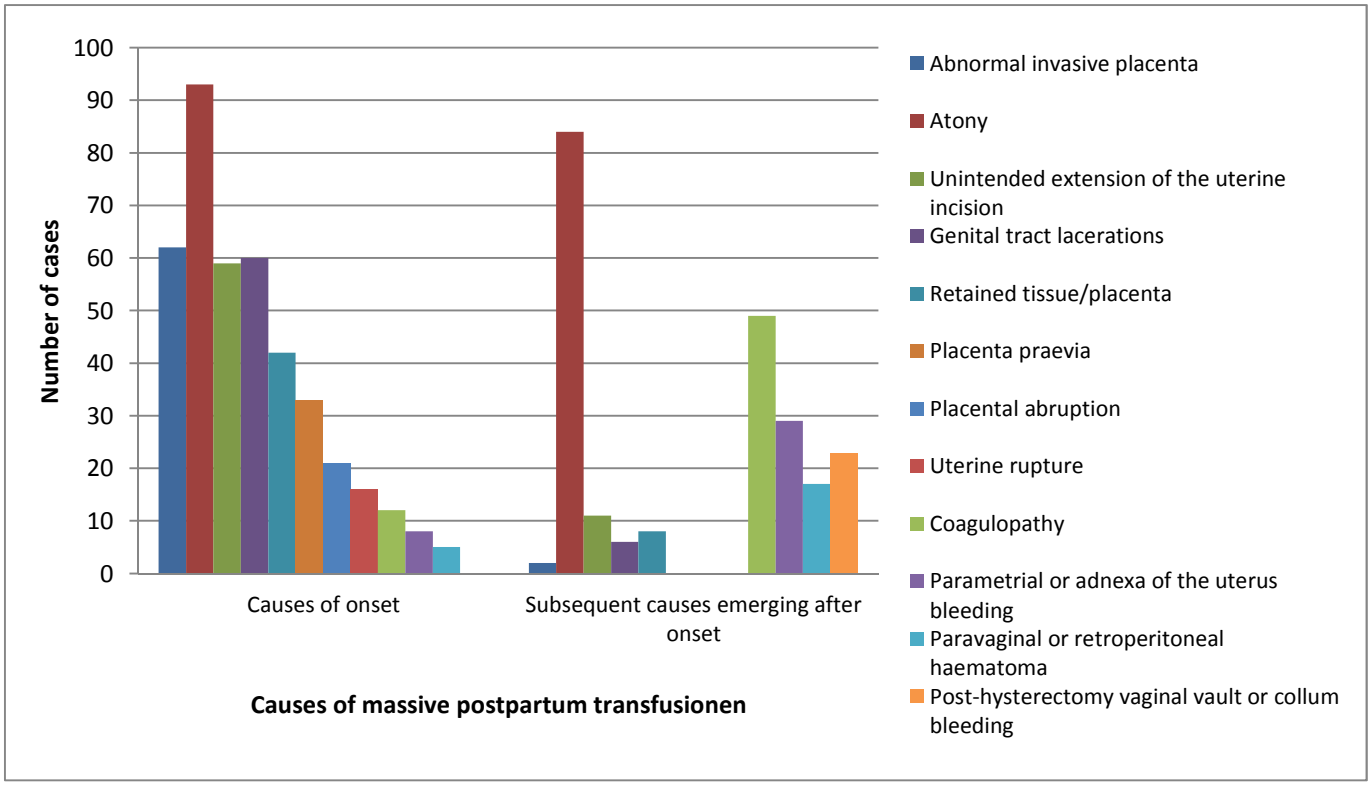


Figure 3: Primary causes of onset and subsequent causes emerging after onset of haemorrhage in women with massive postpartum transfusion.



Hysterectomy

In women with massive postpartum transfusion requiring hysterectomy (Table 3), we found that they were significantly older (p<0.001), had lower gestational age (p =0.012), higher parity (p<0.001), a more frequent history of caesarean section (p=0.002) and a higher rate of AIP (p<0.001) than women not in need of a hysterectomy. There was no difference in the rate of caesarean section. Furthermore women with massive postpartum transfusion requiring hysterectomy had greater blood loss (p<0.001), longer time in surgery (p<0.001), and more frequent need of at least 3 laparotomies (p<0.001). A median of 13 (IQR 10-19) RBC units were given before or during hysterectomy (Figure 4), and we found that women with massive postpartum transfusion requiring hysterectomy received higher volumes of RBCs (p<0.001), FFP (p<0.001), PLTs (p<0.001), and recombinant factor VIIa (p=0.002) compared with women not requiring hysterectomy (Table 3). There was no difference in the mean FFP:RBC ratio before the start of surgery leading to haemostasis, but patients requiring hysterectomy in cases of massive postpartum transfusion had received a higher mean FFP:RBC ratio (p=0.010) by the time of haemostasis. Furthermore, we found that women with massive postpartum transfusion requiring hysterectomy had received more units of RBCs and FFP before receiving their first PLT transfusion than women not requiring a hysterectomy (p=0.006), (Table 3).

Not all of the 128 hysterectomies performed led to haemostasis, as 38 (30%) were in need of further surgical management to control bleeding. These women had greater blood loss and received higher volumes of RBCs, FFP, PLTs and recombinant factor VIIa than patients where hysterectomy led to haemostasis (transfusions shown in Figure 4). The 38 women in need of further surgery had a variety of procedures performed to obtain haemostasis, often in combination. A total of 23 (61%) had tamponade of the cervical canal, sutures to the vaginal vault or sutures to/removal of the cervix performed, five (13%) had abdominal packing, nine (24%) had sutures to/removal of the ovary/adnexa, three (8%) had suturing of the bladder, two (5%) had ligation of either the uterine or epigastric artery, and one (3%) needed a splenectomy. In some cases it was unclear what exactly led to haemostasis, but 22 women (58%) had coagulopathy at some point during or after surgery (data not shown).

Discussion

Main findings

The incidence of massive transfusions of ≥10 RBCs in relation to PPH was 0.5 per 1,000 deliveries. Two of the eight patients with cardiac arrest died. The main causes of massive postpartum transfusion were atony, AIP, unintended extension of the uterine incision, placenta praevia and retained tissue/placenta, requiring a variety of procedures to obtain haemostasis, with hysterectomy being the most commonly performed. Of the 128 hysterectomies performed only 70% resulted in haemostasis. The mean FFP:RBC ratio in the whole cohort was 0.48 at time of haemostasis, higher in cases requiring hysterectomy, and we found that women with massive postpartum transfusion requiring hysterectomy received significantly more units of RBCs before their first PLT transfusion.

Figure 4: Median number of transfusions before, during and after hysterectomy, with whiskers indicating IQR. (RBC=red blood cell, FFP=fresh frozen plasma, PLT=platelets).

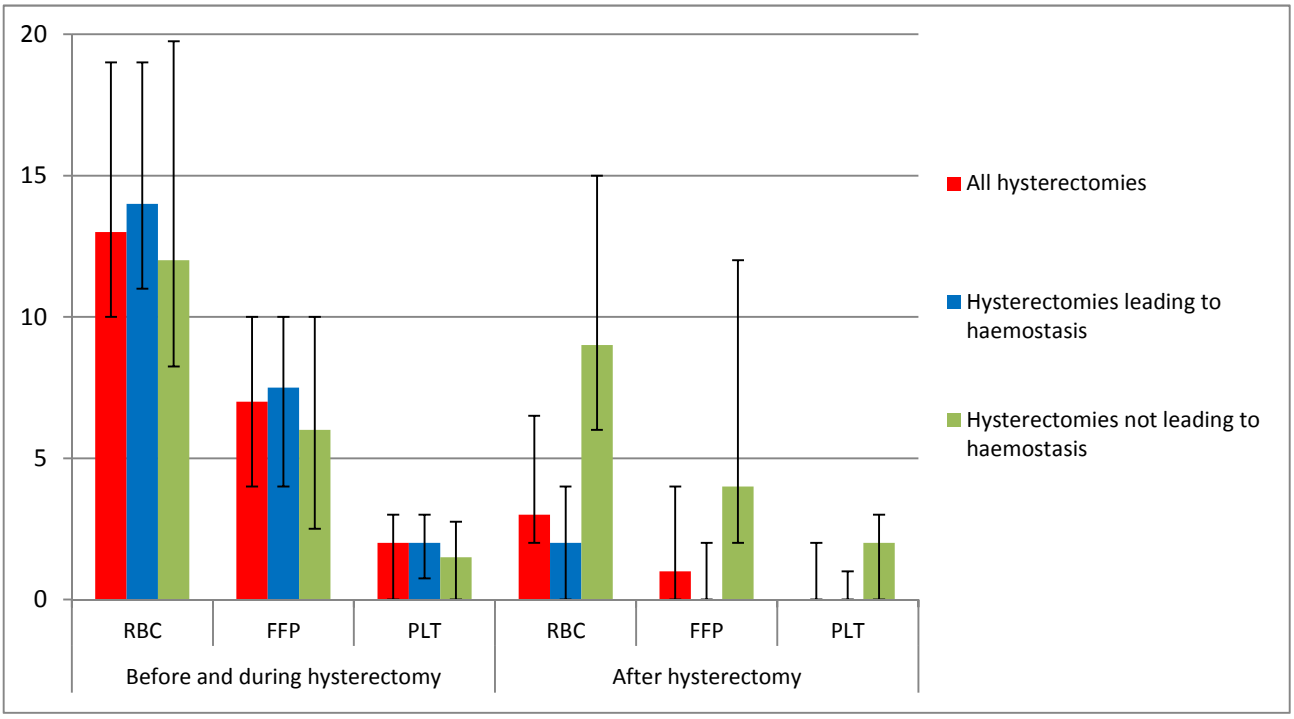


Table 3: Characteristics of women with massive postpartum transfusion with and without hysterectomy. Comparison by univariate logistic regression analysis. (proportions and percent (%) are depicted for categorical data, means and standard deviations (SD) for normally distributed data and medians and interquartile ranges (IQR) for non-normally distributed data**).**

Characteristics	Hysterectomy, n=128 n (%), mean ± SD, median [IQR]	No hysterectomy, n=117 n (%), mean ± SD, median [IQR]	p
Maternal and transfusion characteristics			
Maternal age	34.0 ± 4.6	31.3 ± 5.1	**< 0.001**
Gestational age	266 ± 23.9	274 ± 24.1	**0.012**
Parity	2.4 ± 1.2	1.6 ± 0.8	**< 0.001**
Previous caesarean section	50 (39.1)	24 (20.5)	**0.002**
Abnormal invasive placenta	49 (38.3)	15 (12.8)	**< 0.001**
Caesarean section	86 (67.2)	77 (65.8)	0.82
Characteristics of PPH and treatment			
Number of RBCs before first FFP	6 (4-8)	5 (4-8)	0.95
Number of RBCs before first PLT	10 (6-13)	8 (5-10)	**0.006**
Number of FFP before first PLT	4 (2-5)	2 (1-4)	**0.025**
Mean FFP:RBC ratio before start of surgery leading to haemostasis	0.34±0.31	0.29±0.28	0.249
Total number of RBCs at haemostasis	16 (11-21)	11 (9-14)	**< 0.001**
Total number of FFP at haemostasis	8 (4-12)	4 (2-8)	**< 0.001**
Total number of PLTs at haemostasis	2 (1-4)	1 (0-2)	**< 0.001**
Mean FFP:RBC ratio at haemostasis	0.52±0.21	0.44±0.24	**0.010**
Women receiving recombinant factor VIIa	28 (23.0)	9 (7.9)	**0.002**
Need of ≥3 laparotomies	24 (18.8)	5 (4.3)	**< 0.001**
Total time in surgery	3:50 (3:00-5:10)	2:45 (2:05-3:39)	**< 0.001**
Complications			
Hospitalization time (days)	10 (7-18)	8 (6-12)	**< 0.001**
Total blood loss (L)	9.3 (7.5-1.2)	7.0 (5.5-8.0)	**< 0.001**
Cardiac arrest	5 (3.9)	3 (2.6)	0.56
Death	1	1	-

PPH = postpartum haemorrhage, RBC = Red Blood Cell, FFP = Fresh Frozen Plasma, PLT = Platelet. ICU = Intensive Care Unit. Bold indicates significant p<0.005.

Strengths and limitations

Using a combination of two national registries with known high validity and only excluding 6 cases due to missing patient charts we were able to obtain high external validity.[20] After review of patient charts we were able to exclude cases where blood transfusions were given in close proximity of childbirth but due to non-obstetrical causes. We used a standardized abstraction form and only used one abstractor that went through all patient charts, thereby minimizing interrater variability. We were not affected by attrition bias as none of the data extracted relied on follow-up visits.[21] By choosing a cohort that comprises the known definition of massive transfusion of at least 10 units of RBC, our data becomes more comparable to transfusion studies in other fields. Even though we do not have data regarding timing of transfusions given prior to the decision to perform hysterectomy, the fact that the majority of blood transfusions were given before or during hysterectomy supports the notion that hysterectomy was performed due to severe haemorrhage. In comparison if we had included all women with peripartum hysterectomy we would have included all hysterectomies performed due to various other causes than severe haemorrhage.

Descriptive studies do have limitations. We can only determine association and not causation, since treatment decisions might be confounded by indication e.g. the decision to perform hysterectomy could be influenced by failed attempts to obtain haemostasis by using FFP and PLTs. Furthermore interpretation of effects of treatments strategies becomes difficult as multiple interventions are done simultaneously and without exact documentation of timing. Some of the data in our cohort are over 10 years old, covering a time where much has changed in the regard to transfusion protocols and collaboration between specialties, why maybe not all information gained is relevant in today's obstetric practice.

Interpretation

Women in need of massive postpartum transfusions are very rare and constitute those where usual treatment for PPH has failed or been inadequate. As the rate of PPH in the developed world is 3-8%[22], there are potentially approximately 26,753 deliveries (5.5% of 486,431, see figure 1) with PPH managed without the need for massive transfusion in this cohort. The present study only describes these extreme cases and the different treatment strategies attempted to gain haemostasis.

Similar studies on severe PPH generally assign each patient a single cause relating to the traditional classification of atony, trauma, retained tissue or coagulopathies, without differentiating between causes of onset of haemorrhage and subsequent causes arising later on.[6,23–25] Atony plays a role in the majority of cases with severe PPH, but we found that it often develops secondary to or in combination with other causes. This differentiation makes it hard to compare with other studies that only state the primary cause of aetiology of severe PPH e.g. atony 21-62% and abnormal placentation 12-55%.[6,25–27].

There is only one other study on massive transfusion in obstetrics that uses the generally accepted definition of ≥10 RBC transfusions.[6] They found a similar incidence of 0.6 per 1,000 deliveries from 1998-2007 in the US. Unfortunately they did not include any data regarding FFP, PLTs or other pro-haemostatic treatment. We found a higher FFP:RBC ratio at the time of haemostasis in women with massive postpartum transfusion requiring hysterectomy, but the difference between the ratios of 0.08 is of minimal clinical significance, and could be due to the longer total time in surgery giving the anaesthetist more time to consider a balanced transfusion strategy.[28] Likewise we found larger volumes of RBCs, FFP and PLTs transfused in the hysterectomy group, most likely due to larger blood loss, and not necessarily as others conclude – a risk factor for hysterectomy.[4] The effect of the clinical evaluation leading to the surgical plan is difficult to capture in studies like this. In cases of massive haemorrhage where surgical control is deemed difficult, or FFP is not easily available, early hysterectomy can be an option that is considered to control bleeding. In comparison, a recent study from the UK including 179 women with PPH and ≥8 RBC transfusions, 45% required hysterectomy and the median FFP:RBC ratio was 0.5.[29] This is a lower ratio than the last 4 years of our cohort, but could be due to the lower threshold for inclusion. Unfortunately it was not possible to investigate the exact number of transfusions given at the time the hysterectomy decision was made, and we are therefore not capable of estimating the FFP:RBC ratio at this exact point of time.

Severe complications defined by end organ dysfunction were seen in 44.5% of our cohort. In a study from Ireland from 2004-2007 end organ dysfunction was found in 19% of patients with transfusion of ≥5 RBCs.[4] The only other study of ≥10 RBCs did not define end organ dysfunction, but found 33% had acute renal failure, mechanical ventilation or died. We cannot conclude whether our cohort had more severe complications due to the different definitions, but there is no doubt that morbidity is very high in women with massive postpartum transfusions.

The rate of hysterectomy also varies depending on the inclusion criteria, as a lower transfusion threshold means that aspects other than bleeding can influence the decision to perform a hysterectomy. In studies including women with transfusion of ≥5-10 RBCs hysterectomy was required in 24-45% [4,6,25] compared to 52% in our study. The success rate of hysterectomy was 70%, which is comparable to similar studies, where the success rates vary from 50-85%.[23,30–32] This is quite controversial as obstetricians and anaesthetists alike seem to consider a hysterectomy as the last option more or less guaranteed to secure haemostasis.

Women requiring a hysterectomy received significantly more RBC transfusions before their first PLT transfusion than women not requiring hysterectomy. This might indicate that a transfusion strategy with early PLT transfusions could be helpful in prevention of hysterectomy in cases of massive postpartum transfusion. A randomised controlled study comparing different transfusion strategies would have been preferred, but is nearly impossible in such a rare condition. Our findings are in alignment with findings from trauma and non-trauma where the importance of early PLT transfusion has been shown in cases of massive transfusion.[12–15] A recent study of PLT requirements during severe PPH (median PPH 8,000 ml) found no need for PLT transfusion in cases of PPH <5000 ml or cases without antenatal thrombocytopenia or consumptive coagulopathy. This led to the conclusion that no evidence exists to support the use of early fixed-rate PLT transfusions in uncomplicated severe PPH.[33] However, this study did not include any measures of outcome of the women included and therefore the effect of restrictive PLT transfusions cannot be measured. Furthermore, in an acute scenario with a severely bleeding patient it is difficult to know if consumptive coagulopathy has developed or not. Even if the treatment is uncalled for in some cases, PLTs might still play an important role in the most severe cases, and focus should be on the best timing and monitoring of haemostasis to evaluate further goal-directed use of PLT transfusions.[34]

Conclusion

We found a high incidence of severe morbidity in women requiring massive postpartum transfusion with 52% requiring hysterectomy. The main causes of onset of PPH were atony, AIP and genital tract lacerations, with atony often seen emerging after onset of PPH and often in combination with other causes. Hysterectomy is generally considered last treatment option guaranteed to secure haemostasis, but only 70% of the hysterectomies in our cohort resulted in haemostasis. Women treated with a hysterectomy had increased blood loss and increased transfusion requirements at the time of haemostasis compared to women not requiring hysterectomy.

Acknowledgements

We would like to thank Tobias W Klausen for his contribution to the statistical analyses.

Disclosure of interests

HE, AW, JLR, AA, AMM, JL, JAS and JS have no conflicts of interest or financial ties to disclose.

Contribution to authorship

AW, JS and HE developed the research question. JLR, AW and JS provided supervision throughout the study. HE carried out the data analyses and wrote the manuscript. AW, JLR, AA, AMM, JL, JAS and JS contributed with interpretation of results and revision of the manuscript.

Funding

This study received funding from The Department of Obstetrics and Gynaecology, Herlev Hospital, Denmark and The Danish Society of Anaesthesiology and Intensive Care Medicine.

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Table S1: Additional characteristics of women with massive postpartum transfusion (proportions and percent (%) are depicted for categorical data, means and standard deviations (SD) for normally distributed data and medians and inter-quartile ranges (IQR) for non-normally distributed data**).**

Characteristics	n (%), mean ± SD, median [IQR]
Characteristics of PPH	
Women with secondary PPH (missing n=3)	7 (2.9)
Time from birth to start of haemorrhage (hours:minutes) (missing n=3)	0:00 [-0:07-0:08]
Characteristics of transfusions	
Women receiving preoperative RBCs (missing n=2)	14 (5.8)
Women receiving preoperative FFP (missing n=5)	1 (0.4)
Women receiving preoperative PLTs (missing n=6)	0 (0.0)
Fibrinogen before initiating haemostasis surgery, any	17 (7.0)
Cryoprecipitate before initiating haemostasis surgery, any	4 (1.7)
Recombinat factor VIIa before initiating haemostasis surgery, any	12 (5.0)
Albumin before initiating haemostasis surgery, any	23 (9.6)
Tranexamic acid before initiating haemostasis surgery, any	96 (40.0)
FFP:RBC ratio at end of haemostasis surgery (missing n=7)	0.48±0.23
2001	0.41±0.28
2002	0.45±0.24
2003	0.39±0.19
2004	0.40±0.21
2005	0.45±0.16
2006	0.52±0.23
2007	0.52±0.19
2008	0.55±0.20
2009	0.62±0.29
Total number of RBC (missing n=9)	
Moderate (<15)	95 (40.3)
High (15-20)	84 (35.6)
Massive (>20)	57 (24.2)
Total number of FFP (missing n=10)	8 [5-12]
Total number of PLT (missing n=10)	2 [1-4]
Total crystalloids (ml)	7,000 [5,000-8,500]
Total colloids (ml)	2,000 [1,500-2,500]
Fibrinogen concentrate, any	50 (21.2)
Cryoprecipitate, any	7 (3.0)
Recombinat factor VIIa, any	37 (15.7)
Albumin, any	44 (19.1)
Tranexamic acid, any	161 (68.5)
Complications	
Total blood loss (ml) (missing n=17)	8,000 [6,500-10,375]
<5,000	19 (8.3)
5,000-10,000	152 (66.7)
>10,000	57 (25.0)

Perioperative assistance from other specialty	39 (15.9)
Contact to thrombosis and haemostasis expert	60 (24.5)
Coagulopathy at any time (missing n=2)	84 (34.6)
Transferred to tertiary hospital due to severe postpartum haemorrhage	15 (6.1)
Obstetric interventions	
Women receiving preoperative oxytocin	104 (42.6)
Women receiving preoperative misoprostol	38 (15.6)
Women receiving preoperative methergine	29 (11.9)
Women receiving preoperative prostinfenem	6 (2.5)
1 laparotomy	81 (33.0)
2 laparotomies	105 (43.0)
≥3 laparotomies	29 (11.8)
Total number of surgeries	2 [2-3]
Women requiring hysterectomy	128 (52.2)
Women requiring further surgery after hysterectomy	28 (19.3)
Hysterectomy at 1st laparotomy	70 (54.5)
Hysterectomy at 2nd laparotomy	48 (37.5)
Hysterectomy at 3rd laparotomy	8 (6.3)
Hysterectomy at 4th laparotomy	2 (1.6)
Total surgery time (hour:minutes)	3:23 [2:20-4:15]
Timing of interventions	
Time from start of haemorrhaging to first surgery (hours:minutes)(missing n=2)	0:15 [-0:06-1:15]
Vaginal delivery (hours:minutes) (n=81)	1:10 [0:41-2:37]
Caesarean section (hours:minutes) (n=162)	-0:04 [-0:12-0:15]
Time from start of haemorrhage to first massive transfusion of RBCs (hours:minutes)	2:00 [0:49-3:49]
Time from start of haemorrhage to first FFP (hours:minutes)	3:15 [1:49-6:00]
Time from start of haemorrhage to first PLT (hours:minutes)	4:40 [2:27-8:17]
Time from start of first FFP to haemostasis (hours:minutes)	3:10 [1:30-8:04]
Time from start of first PLT to haemostasis (hours:minutes)	1:45 [0:32-5:49]

PPH = postpartum haemorrhage, RBC = Red Blood Cell, FFP = Fresh Frozen Plasma, PLT = Platelet.

Causes and predictors of postpartum blood loss: a cohort study

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Abstract

Objectives

To describe the distribution of causes of postpartum blood loss depending on the cut-off used to define postpartum haemorrhage, and to describe the association between quantity of blood loss, duration of the third stage of labour, retained placenta and other risk factors.

Design

Cohort study.

Setting

Two Danish maternity-units.

Population

All vaginal deliveries between 1 January 2009 and 31 December 2013 (n=43,357).

Methods

Univariate and multivariate linear regression analyses.

Main Outcome Measures

Postpartum blood loss.

Results

The distribution of causes depended on the cut-off used to define postpartum haemorrhage. In cases of blood loss ≥500 ml retained placenta constituted 12%, lacerations 57% and other causes including atony 31%. In blood loss ≥1,000 ml retained placenta constituted 34%, lacerations 44% and other causes including atony 22%. In blood loss ≥1,500 ml retained placenta constituted 47%, lacerations 37% and other causes including atony 16%. In blood loss ≥2,000 ml retained placenta constituted 53%, lacerations 34% and other causes including atony 14%. Retained placenta was a strong predictor of quantity of blood loss. Duration of the third stage of labour was a weak predictor and the predictive power was reduced further in the multivariate analysis when including retained placenta in the model.

Conclusions

There was a significant difference in distribution of causes depending on the cut-off used to define postpartum haemorrhage. The predictive power of the duration of the third stage of labour was diminished by the influence of retained placenta.

Introduction

It is well-known worldwide that the leading direct cause of maternal morbidity and mortality in both developing and developed regions is postpartum haemorrhage (PPH).[1] In recent years the awareness towards PPH has increased further due to a rising incidence in developed regions.[2–4] This rise has not only been seen in cases of mild PPH, but also in more severe cases.[5,6] The major cause of PPH has typically been attributed to lack of tone in the uterus, i.e. atony, with the remainder of causes associated with either trauma to the birth canal, retained tissue or coagulopathy.[7] However, the cut-off used to define PPH and the distribution of causes varies between studies, leaving us unsure whether it is the different cohorts or the different definitions that cause the variation in distribution.[5,8–10]

Prolonged duration of the third stage of labour has been identified as one of the risk factors for PPH, leading to most national guidelines recommending active management of the third stage of labour and manual removal of the placenta if this exceeds 30 minutes or if there is bleeding.[5,11–13] Recent studies have, however, reported that the increased risk of PPH already exists if the third stage of labour exceeds 15 to 20 minutes, suggesting that manual removal of the placenta should be performed early on regardless of blood loos.[14–16] Up to 90% of placentas are delivered within 10 minutes,[15,17] with the majority of remaining cases having a prolonged duration of the third stage of labour due to a retained placenta that is entrapped, adherent or even abnormal invasive (AIP).[18,19] These disorders are all risk factors for PPH and often require manual removal of the placenta irrespective of the length of the third stage of labour. However, they are difficult to diagnose before manual removal has taken place.[19] The question is therefore whether some placentas removed manually could have been delivered spontaneously without an increased risk of PPH regardless of the length of the third stage of labour.

In the present study we analysed data from a large regional birth cohort to investigate whether the distribution of causes of postpartum blood loss depended on the cut-off used to define PPH. Furthermore, we investigate the association between the duration of the third stage of labour, a retained placenta, other risk factors, and quantity of postpartum blood loss.

Methods

Study population

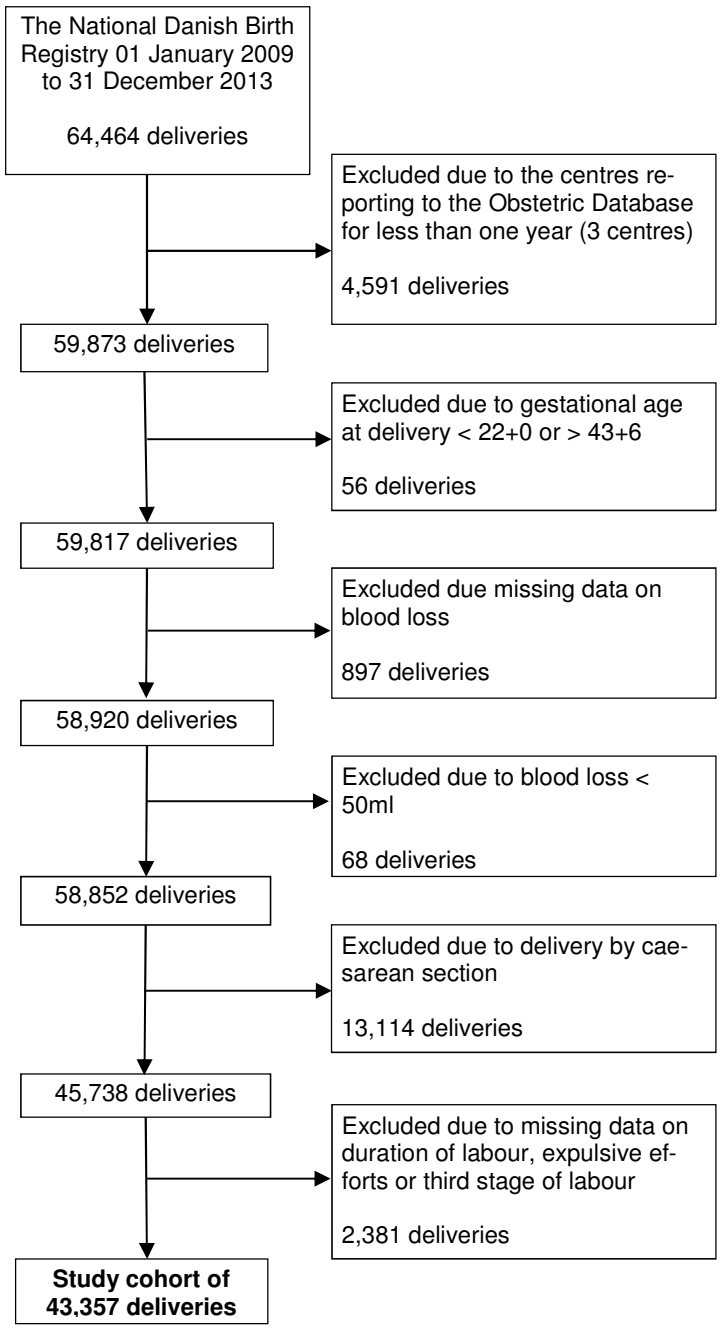
This retrospective cohort study is based on data from the Copenhagen Obstetric Database where all births in the Capital Region of Copenhagen are registered. Data from 01 January 2009 until 31 December 2013 were retrieved. We acquired permission from the Danish Data Protection Agency (Journal nr.2012-58-0004). No permission was required from the Danish Ethics Committee according to Danish legislation.

The Obstetric Database includes data on maternal demographics, parity, labour, and delivery, and holds a high internal validity with kappa coefficients from 0.7-1.0.[20] Midwives register baseline data and interventions both during and after labour, while obstetricians or senior midwives supply specialist diagnosis after the patients have been discharged from hospital.[20] Estimations of blood loss at the hospitals are based on visual estimation, weighing of pads, collector bags, and collection during peripartum surgery. The Danish national guideline recommends routine administration of 10 IU of oxytocin at delivery and manual removal of the placenta within 30 minutes or immediately if there is active bleeding.[11] Inclusion criteria were all vaginal births (singleton and multiple pregnancies) from 22 to 43 weeks of gestation, as the majority of cases with GA >43+6 were interpreted as faulty registration. All cases with missing data on blood loss (897 deliveries), or postpartum blood loss lower than 50 ml (68 deliveries) were excluded, due to the assumption that all women bleed at least 50 ml. Due to some hospitals only reporting to the registry for less than one year of the study period we excluded these hospitals (3 centres/4,591 deliveries).

Variables

The following variables from the database were included in this study: maternal age, parity, previous caesarean section, singleton or multiple pregnancy, hypertensive disorders of the mother, antepartum haemorrhage, placenta praevia, abnormal invasive placenta, presentation of the foetus, amniotic fluid abnormalities, preterm premature rupture of membranes (PPROM), premature rupture of membranes (PROM), date and time of delivery, hospital, gestational age at delivery, induction of labour, augmentation of labour, fever during labour, mode of delivery, episiotomy, genital tract lacerations, epidural analgesia, uterine rupture, placental abruption, shoulder dystocia, uterine inversion, quantity of postpartum blood loss (ml), duration of labour (min), duration of expulsive efforts (min), time of delivery or removal of the placenta, retained placenta and manual removal of tissue or placenta. The diagnosis "retained placenta" is not used if the placenta is delivered spontaneously, regardless of the length of the third stage of labour. Abnormal invasive placenta, retained placenta and manual removal of tissue or placenta were grouped together as "retained placenta". Cases with missing or invalid durations of labour, expulsive efforts and third stage of labour were excluded from the linear regression model. All cases were given a single cause of postpartum blood loss: 1) "Retained placenta or tissue" for all cases involving one of the above mentioned diagnoses; 2) "Lacerations" for all cases without retained placenta or tissue and with a minimum of grade two lacerations; 3) "Other including atony" for all cases with none of the above. We were not able to retrieve sufficient data on previous PPH, BMI and birthweight. Outcome of quantity of postpartum blood loss was not only analysed as a continuous variable but also using four different definitions of PPH with cut-offs of: ≥500 ml, ≥1,000 ml, ≥1,500 ml, and ≥2000 ml. Duration of the third stage of labour was calculated from the time of delivery of the neonate until the time of delivery of placenta - either spontaneously or by manual removal.

Figure 1. Flow chart of excluded deliveries for the analysis of risk factors for quantity of postpartum blood loss.



Statistical analysis
All data analyses were carried out using SPSS 22.0 (SPSS, Chicago, IL, USA). Quantity of postpartum blood loss and durations are presented as medians and interquartile ranges (IQR) due to their non-parametric distribution. Quantity of postpartum blood loss was logarithmic transformed (log10) due to substantially skewed distribution. Risk factors for quantity of postpartum blood loss were analysed using univariate and multivariate linear regression analyses presented as β-coefficients and 95% Confidence Intervals (CI). Due to the logarithmic transformation of quantity of postpartum blood loss, β-values need to be back-transformed by raising 10 to the power of each value and subtracting one (10^{x}-1); and can then be interpreted as percent change in the predicted quantity of postpartum blood loss. Thereby the predicted mean blood loss can be calculated for each risk factor using the constant β and the percent change. The predicted mean blood loss for a combination of risk factors can be calculated by adding the β coefficient for each risk factor to the β coefficient of the constant, before "back-transforming".

All variables extracted from the database were included in the multivariate linear regression model stepwise with model 1 only including variables known before and during pregnancy, model 2 including variables arising during labour and delivery and finally model 3 including a retained placenta. All variables were tested for correlation using Pearson's correlation in the linear regression analysis, and there were no significant correlations between any two variables. Chi^2 test was used to analyse postpartum blood loss as a categorical variable.

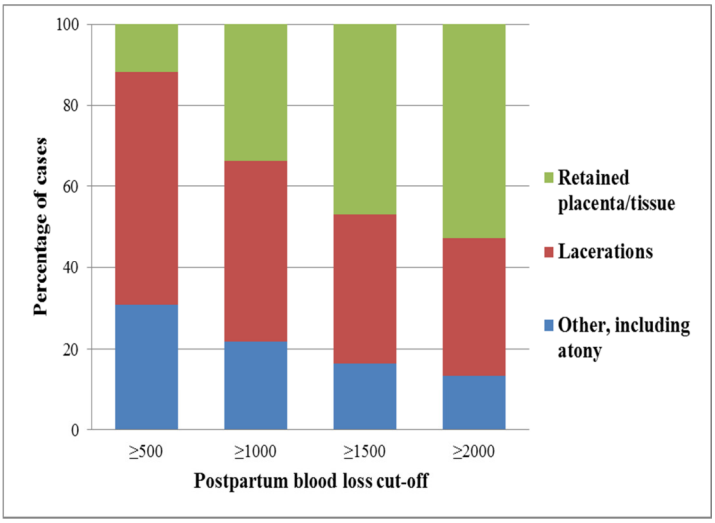
Data are presented as proportions and percent (%), means and standard deviations (SD), or medians and interquartile ranges (IQR) for normally and non-normally distributed data. A two-tailed p-value of less than 0.05 was considered statistically significant.

Results
We identified 43,357 vaginal deliveries from the Copenhagen Obstetric Database from 2009 till 2013 (Figure 1). The majority of deliveries were by nulliparous women (53.9%) between 20 and 34 years of age (75.6%) without any hypertensive disorders (95.5%). A total of 92.2% gave birth at term with a median duration of the third stage of labour of 8 minutes [IQR 5-12] and a median blood loss of 300 ml [IQR 200-400] (Table S1).

Causes of postpartum blood loss
Postpartum blood loss of ≥500 ml was seen in 7541 women (17% of all vaginal deliveries) of which 12% (n=886) were caused by retained placenta, 37% (n=2780) were caused by lacerations and 51% (n=3848) had other causes including atony (Figure 2). This distribution changed significantly the higher the cut-off for postpartum blood loss, with 53% (n=288) caused by retained placenta, 24% (n=133) caused by lacerations and 23% (n=125) having other causes including atony in women with a postpartum blood loss of ≥2000 ml (n=546, 1%) (Figure 2).

Prediction of quantity of postpartum blood loss
Duration of the third stage of labour was identified as a significant but weak predictor of quantity of postpartum blood loss in the univariate linear regression analysis β=0.0046 (p<0.005), equivalent to a predicted increase in postpartum blood loss of 10.7% for every prolonged 10 minutes (Table 1). The multivariate linear regression analysis predicted the mean blood loss to 224 ml (95% CI: 219 to 229, β=2.350) for a delivery without the presence of any of the included risk factors. The predictive power of the duration of the third stage of labour remained unchanged after adjusting for the majority of variables in the multivariate analysis (β=0.004, p<0.005) (Table 1, model 2). However, the effect of duration of the third stage of labour decreased substantially after inclusion of retained placenta in the model to β=0.001 (95% CI: 0.001 to 0.001), equivalent to a predicted increase in postpartum blood loss of 2.9% for every 10 minutes (Table 1, model 3). A full list of all variables investigated in the univariate and multivariate analyses can be seen in Table S1 and S2.

Figure 2. Distribution of causes for different definitions of postpartum blood loss (Number of deliveries in each category: ≥500ml: 7,514, ≥1,000ml: 2,198, ≥1,500ml: 1,113, ≥2,000ml: 546).



We have plotted the median quantity of postpartum blood loss for different time intervals of the third stage of labour in order to illustrate the effect of duration of the third stage of labour (Figure 3). However, as we have identified retained placenta or tissue as a major contributor to a decrease in effect size, we have also stratified the cohort and illustrated them independently. The graph illustrates that for all vaginal births there is a rise in median quantity of blood loss if the length of the third stage exceeds 60 minutes. After stratification, however, there is minimal change in the median blood loss for retained placenta; regardless of the length of the third stage, they all have a median blood loss of approximately 1,500 ml. Likewise, for women with spontaneous delivery of an intact placenta there is no change in the median blood loss (Figure 3).

Figure 3. Median quantity of postpartum blood loss in relation to the duration of the third stage of labour. All vaginal deliveries and also stratified for retained placenta. Whiskers indicate Interquartile Range (Retained placenta = abnormal invasive placenta, retained placenta and manual removal of placenta or tissue, No retained placenta = spontaneous delivery of an intact placenta).

The multivariate linear regression model also identified other predictors of blood loss (Table 1). Uterine rupture had the highest effect on prediction of blood loss in our multivariate linear regression model (β=0.625), but was only seen in two cases in this cohort of vaginal deliveries and with a blood loss of 800 ml and 3,000 ml respectively (Table 1). The remaining variables with the highest significant effect in the multivariate analysis were: retained placenta (β=0.544), cervical lacerations (β=0.497), eclampsia (β=0.254), placenta praevia (β=0.144), 3rd and 4th degree perineal lacerations (β=0.141) and upper vaginal lacerations (β=0.140) (Table 1). Overall the predictors in the final model accounted for 23.2% of the variability in quantity of postpartum blood loss (R^2=0.232). The proportion of cases without any predictors of blood loss was therefore also high, comprising 20.6% (n=8,936) of all vaginal deliveries including women with blood loss <500 ml. However, the proportion of cases without any risk factors decreased significantly the higher cut-off used to define PPH from 11% (n=806) of cases with a postpartum blood loss ≥500 ml, 7% (n=151) of cases ≥1,000 ml, 5% (n=52) of cases ≥1,500 ml and 4% (n=22) of cases ≥2,000 ml.

Discussion

Main findings

In this retrospective cohort study of 43,357 vaginal deliveries, we found that the distribution of causes depended on the cut-off used to define PPH, with a retained placenta playing an increasing role the higher the cut-off. Furthermore, we found that the duration of the third stage of labour was associated with the quantity of postpartum blood loss, but that the effect size of this association diminished after adjusting for retained placenta.

Strengths and Limitations

The main strength of this study is the large cohort size and the known high validity of the Obstetric Database.[20] Furthermore, the data are from two large university hospitals in the capital region of Denmark representative of the Danish population, due to the known substantial sociodemographic and healthcare homogeneity with the remaining four regions of Denmark.[21] Most other studies investigating risk factors for PPH have dichotomised postpartum blood loss into values above or below a certain threshold, thereby facilitating logistic regression analysis and results in odds ratios.[10,13,22–24] Overall, we have identified many of the same risk factors, but interpretation of the results is more challenging. An odds ratio can only describe the odds of blood loss more than e.g. 500 ml compared to less than 500 ml, thereby not differentiating between 550 ml or 2000 ml. By analysing quantity of postpartum blood loss as a continuous outcome, we were able to identify the effect on quantity of each variable.

Limitations of this study are mainly attributed to missing information on some risk factors such as previous PPH, birthweight, and BMI. The few studies that included data on BMI or previous PPH have shown either non-significance or low odds ratios of 1.5-2.2.[25–27] Macrosomia is a well-known risk factor for PPH with odds ratios of 1.4-3.5,[5,13,28] and therefore a possible confounder in our study. We have, however, been able to include shoulder dystocia in our analyses, which is a proxy variable for macrosomia. However, the cohort was sufficient in size to test for all included risk factors without overfitting. The main limitation of this study in regard to the association between the length of the third stage of labour and quantity of postpartum blood loss

Table 1: Univariate and multivariate linear regression analysis of variables for prediction of quantity of postpartum blood loss after vaginal delivery (proportions and percent (%) are depicted for categorical data and medians and interquartile ranges (IQR) for non-normally distributed data).

Variables	N (%) or median [IQR]	Univariate linear regression		Model 2		Model 3		Percent change in outcome (%)**
		β	p-value	β	95% CI	β	95% CI	
Constant				**2.325**	2.315;2.334	**2.350**	2.341;2.360	
Duration of third stage of labour (minutes)	8 [5-12]	**0.0046***	<0.005	**0.004**	0.004;0.004	**0.001**	0.001;0.001	**2.9*****
Before and during pregnancy								
Previous caesarean section	1,649 (3.8)	**0.060**	<0.005	**0.057**	0.044;0.069	**0.054**	0.042;0.065	**13.2**
Hypertensive disorders								
None	41,419 (95.5)	Ref		Ref		Ref		
Gestational hypertension	783 (1.8)	**0.050**	<0.005	0.016	0.000;0.033	0.015	-0.001;0.031	
Mild/moderate preeclampsia	957 (2.2)	**0.073**	<0.005	**0.026**	0.011;0.042	**0.020**	0.005;0.035	**4.7**
Severe preeclampsia	143 (0.3)	**0.151**	<0.005	**0.091**	0.053;0.130	**0.081**	0.045;0.118	**20.5**
Eclampsia	4 (0.0)	0.240	0.057	**0.242**	0.015;0.470	**0.254**	0.037;0.472	**79.5**
HELLP	51 (0.1)	**0.140**	<0.005	**0.087**	0.023;0.150	0.054	-0.007;0.115	
During labour and delivery								
Induction of labour								
None	31,770 (73.3)	Ref		Ref		Ref		
Medical	8,739 (20.2)	**0.044**	<0.005	**0.021**	0.015;0.027	**0.018**	0.012;0.024	**4.2**
Artificial rupture of membranes	4,687 (10.8)	**0.041**	<0.005	**0.025**	0.018;0.025	**0.025**	0.017;0.032	**5.9**
Balloon dilation	74 (0.2)	0.048	0.105	-0.027	-0.081;0.027	-0.018	-0.069;0.033	
Augmentation of labour	14,382 (33.2)	**0.080**	<0.005	**0.025**	0.019;0.031	**0.024**	0.019;0.030	**5.7**
Fever during labour	1,186 (2.7)	**0.120**	<0.005	**0.037**	0.023;0.051	**0.035**	0.022;0.048	**8.4**
Episiotomy	1,793 (4.1)	**0.086**	<0.005	**0.073**	0.062;0.085	**0.071**	0.060;0.082	**17.8**
Outlet operative vaginal delivery	2,871 (6.6)	**0.085**	<0.005	0.009	0.000;0.019	0.006	-0.003;0.016	
Low/mid cavity operative vaginal delivery	2,206 (5.1)	**0.120**	<0.005	**0.031**	0.020:0.041	**0.026**	0.016;0.036	**6.2**
Genital tract lacerations								
None	19,714 (45.5)	Ref		Ref		Ref		
Perineal 1st/2nd	20,710 (47.8)	**0.065**	<0.005	**0.055**	0.051;0.060	**0.056**	0.051;0.061	**13.8**
Perineal 3rd/4th	1,610 (3.7)	**0.189**	<0.005	**0.146**	0.134;0.158	**0.141**	0.129;0.153	**38.4**
Upper vaginal	1,137 (2.6)	**0.170**	<0.005	**0.147**	0.133;0.161	**0.140**	0.126;0.153	**38.0**
Cervical	186 (0.4)	**0.700**	<0.005	**0.632**	0.598;0.665	**0.497**	0.465;0.529	**214.1**
Uterine rupture	2 (0.0)	**0.701**	<0.005	**0.630**	0.307;0.953	**0.625**	0.317;0.934	**321.7**
Placental abruption	116 (0.3)	**0.093**	<0.005	**0.092**	0.046;0.137	**0.088**	0.044;0.131	**22.5**
Shoulder dystocia	507 (1.2)	**0.119**	<0.005	**0.081**	0.061;0.102	**0.075**	0.056;0.095	**18.9**
Uterus inversion	5 (0.0)	**0.457**	<0.005	**0.395**	0.191;0.598	**0.024**	0.046;0.434	**5.7**
Postpartum								
Retained placenta	964 (2.2)	**0.642**	<0.005			**0.544**	0.527;0.560	**250.0**

Values are β coefficients and 95% Confidence intervals (CI). Bold β indicates $p < 0.05$. A one minute increase in the three duration variables represents a unit change in log10 of quantity of postpartum blood loss.
Model 2: is a multivariable linear regression of all variables listed in the table apart from retained placenta. In addition adjusted for maternal age, parity, multiple pregnancy, antepartum haemorrhage, non-cephalic presentation, amniotic fluid abnormalities, preterm premature rupture of membranes, premature rupture of membranes, year of delivery, time of day, hospital, induction of labour, mode of delivery, duration of labour, duration of expulsive efforts, and fever during labour. Model 3: All variables in model 2 + retained placenta.
*β coefficient for duration is the change per minute.
**Percent change in outcome: The percentage each variable increases or decreases the predicted blood loss in model 3
***Percent change in the outcome with a 10 min change in duration

relates to the observational design. Women with "retained placenta" is a heterogeneous group, as some women in this group might not have needed manual removal of placenta as many factors can influence this decision. These include availability of obstetricians and operating theatre, but also the severity of bleeding.

Interpretation

This is the first study investigating the change in distribution of causes using different cut-offs to define PPH in the same cohort. We chose to use cut-offs as this gives a different clinical perspective than using bands to group the quantity of PPH. In a clinical setting we are interested in identifying and preventing the most severe cases, thereby is seems relevant to look at all cases with PPH above a certain limit such as above 1,500 ml and not cases with PPH 1,000-1,500 ml or 1,500-2,000 ml.

Other studies have shown changes in the distribution of causes but have only compared PPH with severe PPH.[13,27,28] A study from Israel defined the severity of PPH not by quantity but by different interventions.[23] They found that the rate of retained placenta increased from 0% of women with PPH and no need for intervention to 1.8% of women with PPH in need of revision of the uterus and 4.5% of women with PPH requiring blood transfusions. Similarly, a Dutch study found an increase in retained placenta 7.4% in PPH >500 ml to 25.5% in PPH >1,000 ml.[28] None of the studies assessed the rate of atony, but 49-75% had neither lacerations or retained placenta, which is markedly more than in our study. A study from Turkey analysed the different causes of PPH in women with PPH >500 ml and women with severe PPH (PPH ≥2,000 ml, Haemoglobin decrease ≥2g/dl, transfusion of ≥4 packed red blood cells, haemostatic intervention or death).[27] They found an increase in atony (40.4% to 62.4%), and an increase in retained placenta including abnormal placentation (11.6% to 16.0%), illustrating again that atony played a more substantial role compared to our study. These discrepancies in the role of atony could be explained by how the different studies defined atony. We defined atony only as an exclusion diagnosis, thereby presumably including cases caused by other causes and missing cases caused by atony. Atony as a diagnosis is often used when no other cause is obvious or in cases of PPH, where bleeding from the uterine cavity is misinterpreted as atony, but is in fact bleeding from the placental site due to AIP or coagulopathy developed later on in the course of events.

The minimal effect, the duration of the third stage of labour had on quantity of postpartum blood loss in our study contradicts the majority of other research to date. One of the reasons for these conflicting findings is the role of the retained placenta. Retained placenta is a risk factor often identified after delivery and a known cause for PPH. Apart from uterine rupture, it was the variable with the highest impact on prediction of quantity of blood loss, in accordance with other studies.[3,13,23,28] In our analyses it was comprised of several different diagnoses, most of which were made after delivery (e.g. manual removal of placenta and tissue), which could explain some of the disparities. The majority of studies have not included retained placenta in their analyses and are thereby not comparable.[14,15] However, a large study investigating the risk of postpartum blood transfusions and duration of the third stage of labour excluded all women in need of manual removal, and still found that a third stage longer than 17 minutes was associated with an increased risk of blood transfusion.[17] A Dutch study investigating risk factors for standard (>500 ml) and severe (>1,000 ml) PPH included duration of the third stage of labour (>30 min) and retained placenta in their multivariate logistic regression analysis.[28] They found an odds ratio of 2.61 if the third stage of labour was >30 minutes for PPH >500 ml and 4.90 for PPH >1,000 ml. Interestingly, their multivariate logistic analysis including only 5 other variables alongside retained placenta, decreased the odds ratios from the univariate analysis substantially from 4.07 to 2.61 for PPH >500 ml, and from 11.9 to 4.90 for PPH >1,000 ml, in accordance with our findings. One could argue that retained placenta was the cause of a prolonged third stage, but the data in figure 3 also suggest that a woman could have a prolonged third stage and have spontaneous delivery of an intact placenta, but no increase in blood loss. Therefore, it seems evident to focus on identifying women requiring manual removal, to be able to initiate removal as soon as possible after delivery of the neonate, to prevent further postpartum blood loss. Today up to 50% of abnormal invasive placenta cases can be diagnosed before delivery by ultrasound and magnetic resonance imaging, [29,30] compared to hardly any cases of entrapped or adherent placenta.[18,31] Therefore, identification of all women requiring manual removal is as yet not possible, but could be improved by advances in ultrasound or use of biomarkers that are involved in placental implantation.[32]

Conclusion

There was a significant difference in the distribution of causes depending on the cut-off used to define PPH, with atony found to play a less substantial role in postpartum blood loss than formerly thought. Duration of third stage of labour was a weak predictor of quantity of blood loss, while retained placenta was a much stronger predictor. The predictive power of the duration of the third stage of labour was further reduced when considering the influence of a retained placenta.

Acknowledgements

We would like to thank Steen Rasmussen and Tobias W Klausen for their contribution to data extraction and statistical analyses.

Disclosure of interests

HE, JL, AW, JS and JLR have no conflicts of interest or financial ties to disclose.

Contribution to authorship

HE and JLR developed the research question. JLR provided supervision throughout the study. HE carried out the data analyses and wrote the manuscript. JLR, AW, JS and JL contributed with interpretation of results and revision of the manuscript.

Funding

This study received funding from The Department of Obstetrics and Gynaecology, Herlev Hospital, Denmark.

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Table S1. Univariate linear regression analysis of risk factors for prediction of quantity of postpartum blood loss after vaginal delivery (proportions and percent (%) are depicted for categorical data and medians and interquartile ranges (IQR) for non-normally distributed data).

Variables	**N (%) or median [IQR n=43,357**	**β**	**p-value**	***% change in the outcome compared to the reference predictor variable***
Blood loss (ml)	300 [200-400]			
Before pregnancy				
Maternal age (years)				
<20	292 (0.7)	-0.017	0.252	-
20-34	32,792 (75.6)	Ref		
>34	10,273 (23.7)	-0.005	0.095	-
Parity				
1st	23,359 (53.9)	Ref		
2-3	18,737 (43.2)	-0.065	<0.005	-13.9
4+	1,261 (2.9)	-0.068	<0.005	-14.5
Previous caesarean section	1,649 (3.8)	0.060	<0.005	14.8
During pregnancy				
Foetal number				
1	42,856 (98.8)	Ref		
2-3	501 (1.2)	0.152	<0.005	41.9
Hypertensive disorders				
None	41,419 (95.5)	Ref		
Gestational hypertension	783 (1.8)	0.050	<0.005	12.2
Mild/moderate preeclampsia	957 (2.2)	0.073	<0.005	18.3
Severe preeclampsia	143 (0.3)	0.151	<0.005	41.6
Eclampsia	4 (0.0)	0.240	0.057	-
HELLP	51 (0.1)	0.140	<0.005	38.0
Antepartum haemorrhage				
Before 22 weeks gestation	378 (0.9)	0.047	<0.005	11.4
After 22 weeks gestation	373 (0.9)	0.057	<0.005	14.0
Presentation				
Cephalic	42,438 (97.9)	Ref		
Non-cephalic	919 (2.1)	0.057	<0.005	14.0
During labour and delivery				
Gestational age at delivery (weeks)				
22-32	384 (0.9)	-0.085	<0.005	-17.8
33-36	1,386 (3.2)	-0.023	<0.005	-5.2
37-41	39,962 (92.2)	Ref		
>41	1,625 (3.7)	0.071	<0.005	17.8
Time of day				
08:00-17:59	17,096 (39.4)	Ref		
18:00-07:59	26,261 (60.6)	0.009	<0.005	2.1
Induction of labour				
None	31,770 (73.3)	Ref		
Medical	8,739 (20.2)	0.044	<0.005	10.7
Artificial rupture of membranes	4,687 (10.8)	0.041	<0.005	9.9
Balloon dilation	74 (0.2)	0.048	0.105	-
Augmentation of labour	14,382 (33.2)	0.080	<0.005	20.2
Mode of delivery				
Vaginal delivery	38,257 (88.2)	Ref		
Operative vaginal delivery				
Outlet	2,871 (6.6)	0.085	<0.005	21.6
Low/mid cavity	2,206 (5.1)	0.120	<0.005	31.8
Failed	23 (0.1)	0.174	<0.005	49.3
Episiotomy	1,793 (4.1)	0.086	<0.005	21.9
Genital tract lacerations				
None	19,714 (45.5)	Ref		
Perineal 1st/2nd	20,710 (47.8)	0.065	<0.005	16.1
Perineal 3rd/4th	1,610 (3.7)	0.189	<0.005	54.5

Upper vaginal	1,137 (2.6)	0.170	<0.005	47.9
Cervical	186 (0.4)	0.700	<0.005	401.2
Fever during labour	1,186 (2.7)	0.120	<0.005	31.8
Uterine rupture	2 (0.0)	0.701	<0.005	402.3
Uterine inversion	5 (0.0)	0.457	<0.005	45.7
Shoulder dystocia	507 (1.2)	0.119	<0.005	186.4
Epidural analgesia	11,672 (26.9)	0.059	<0.005	14.6
Retained placenta	964 (2.2)	0.642	<0.005	338.5
Duration of labour (minutes)	487 [287-802]	4.528E-5	<0.005	0.3*
Duration of expulsive efforts (minutes)	20 [9-39]	0.001466	<0.005	1.5**
Duration of third stage of labour (minutes)	8 [5-12]	0.004638	<0.005	10.7**

Values are β coefficients and p-values

** **% change in the outcome with a one hour change in the reference predictor variable***

*** **% change in the outcome with a 10 min change in the reference predictor variable***

BMI: body mass index, HELLP syndrome: Haemolysis, elevated liver enzymes and low platelet count.

Table S2. Multivariate linear regression analysis of all models for prediction of quantity of postpartum blood loss after vaginal delivery (proportions and percent (%) are depicted for categorical data and medians and interquartile ranges (IQR) for non-normally distributed data).

Variables	N (%) or median [IQR]	Model 1		Model 2		Model 3		Percent change in outcome (%)*
		β	95% CI	β	95% CI	β	95% CI	
Constant		**2.412**	2.406;2.419	**2.325**	2.315;2.334	**2.350**	2.341;2.360	
Duration of third stage of labour (minutes)	8 [5-12]	**0.004**	0.004;0.005	**0.004**	0.004;0.004	**0.001**	0.001;0.001	**2.9****
Duration of labour (minutes)	487 [287-802]	**1.724E-5**	0.000;0.000	**8.095E-6**	0.000;0.000	**7.867E-6**	0.000;0.000	**0.1*****
Duration of expulsive efforts (minutes)	20 [9-39]	**0.001**	0.001;0.001	**4.96E-4**	0.000;0.000	**4.68E-4**	0.000;0.001	**1.1****
Before and during pregnancy								
Previous caesarean section	1,649 (3.8)	**0.081**	0.069;0.094	**0.057**	0.044;0.069	**0.054**	0.042;0.065	**13.2**
Hypertensive disorders								
None	41,419 (95.5)	Ref						
Gestational hypertension	783 (1.8)	**0.039**	0.022;0.056	0.016	0.000;0.033	0.015	-0.001;0.031	
Mild/moderate preeclampsia	957 (2.2)	**0.057**	0.042;0.073	**0.026**	0.011;0.042	**0.020**	0.005;0.035	**4.7**
Severe preeclampsia	143 (0.3)	**0.128**	0.089;0.168	**0.091**	0.053;0.130	**0.081**	0.045;0.118	**20.5**
Eclampsia	4 (0.0)	**0.254**	0.018;0.490	**0.242**	0.015;0.470	**0.254**	0.037;0.472	**79.5**
HELLP	51 (0.1)	**0.116**	0.050;0.183	**0.087**	0.023;0.150	0.054	-0.007;0.115	
During labour and delivery								
Gestational age at delivery (weeks)								
22-32	384 (0.9)			**-0.078**	-0.104;-0.053	**-0.096**	-0.120;-0.071	**-19.8**
33-36	1,386 (3.2)			**-0.032**	-0.048;-0.016	**-0.031**	-0.047;-0.016	**-6.9**
37-41	39,962 (92.2)			Ref				
>41	1,625 (3.7)			**0.036**	0.024;0.048	**0.035**	0.023;0.046	**8.4**
Induction of labour								
None	31,770 (73.3)			Ref				
Medical	8,739 (20.2)			**0.021**	0.015;0.027	**0.018**	0.012;0.024	**4.2**
Artificial rupture of membranes	4,687 (10.8)			**0.025**	0.018;0.025	**0.025**	0.017;0.032	**5.9**
Balloon dilation	74 (0.2)			-0.027	-0.081;0.027	-0.018	-0.069;0.033	
Augmentation of labour	14,382 (33.2)			**0.025**	0.019;0.031	**0.024**	0.019;0.030	**5.7**

Fever during labour	1,186 (2.7)			**0.037**	0.023;0.051	**0.035**	0.022;0.048	**8.4**
Episiotomy	1,793 (4.1)			**0.073**	0.062;0.085	**0.071**	0.060;0.082	**17.8**
Outlet operative vaginal delivery	2,871 (6.6)			0.009	0.000;0.019	0.006	-0.003;0.016	
Low/mid cavity operative vaginal delivery	2,206 (5.1)			**0.031**	0.020:0.041	**0.026**	0.016;0.036	**6.2**
Genital tract lacerations								
None	19,714 (45.5)			Ref				
Perineal 1st/2nd	20,710 (47.8)			**0.055**	0.051;0.060	**0.056**	0.051;0.061	**13.8**
Perineal 3rd/4th	1,610 (3.7)			**0.146**	0.134;0.158	**0.141**	0.129;0.153	**38.4**
Upper vaginal	1,137 (2.6)			**0.147**	0.133;0.161	**0.140**	0.126;0.153	**38.0**
Cervical	186 (0.4)			**0.632**	0.598;0.665	**0.497**	0.465;0.529	**214.1**
Epidural analgesia	11,672 (26.9)			**0.009**	0.003;0.015	**0.010**	0.004;0.016	**2.4**
Uterine rupture	2 (0.0)			**0.630**	0.307;0.953	**0.625**	0.317;0.934	**321.7**
Placental abruption	116 (0.3)			**0.092**	0.046;0.137	**0.088**	0.044;0.131	**22.5**
Shoulder dystocia	507 (1.2)			**0.081**	0.061;0.102	**0.075**	0.056;0.095	**18.9**
Uterine inversion	5 (0.0)			**0.395**	0.191;0.598	**0.024**	0.046;0.434	**5.7**
Postpartum								
Retained placenta	964 (2.2)					**0.544**	0.527;0.560	**250.0**

Values are β coefficients and 95% Confidence intervals (CI). Bold β indicates $p < 0.05$. A one minute increase in duration of the third stage of labour represents a unit change in log10 of quantity of postpartum blood loss.

Model 1 is a multivariate linear regression of the durations and variables identified before or during pregnancy entered simultaneously, in addition adjusted for maternal age, parity, multiple pregnancy, antepartum haemorrhage, non-cephalic presentation, amniotic fluid abnormalities, preterm premature rupture of membranes and premature rupture of membranes.

Model 2: All variables in model 1 + labour and delivery risk factors, adjusted for year of delivery, time of day, hospital, induction of labour, mode of delivery and fever during labour.

Model 3: All variables in model 2 + retained placenta.

*Percent change in outcome: The percentage each risk factor either increases or decreases the predicted median blood loss in model 3.

**Percent change in the outcome with a 10 min change in duration.

*** Percent change in the outcome with a 60 min change in duration.