Acute obstetric coagulopathy during postpartum haemorrhage is caused by hyperfibrinolysis and dysfibrinogenaemia: an observational cohort study

Running title: Postpartum haemorrhage associated coagulopathy

Lucy de Lloyd L*, Peter V Jenkins†,§, Sarah F Bell*, Nicola J Mutch¶, Julia Freyer Martins Pereira*, Pilar M Badenes**, Donna James††, Anouk Ridgeway††, Leon Cohen*, Thomas Roberts*, Victoria Field*, Rachel E Collis*, Peter W Collins†,§

* Department of Anaestetics and Pain Control, Cardiff and Vale University Health Board, Heath Park Cardiff, UK

† Department of Haematology Cardiff and Vale University Health Board, Heath Park Cardiff, UK

§ Institite of Infection and Immunity, School of Medicine, Cardiff University, UK

¶ Aberdeen Cardiovascular & Diabetes Centre, School of Medicine, Medical Sciences and Nutrition, University of Aberdeen, AB25 2ZD.

** Hospital Universitario y Politécnico, Valencia, Spain.

†† Department of Obstetrics and Gynaecology, Cardiff and Vale University Health Board, Heath Park Cardiff, UK

Corresponding author: Peter Collins,

Address: Haemophilia Centre, Cardif and Vale University Health Board, Heath Park Cardiff, CF14 4XN.

Email: peter.collins@wales.nhs.uk

Telephone: +44 02920744144

Essentials

Haemostatic failure may exacerbate postpartum haemorrhage but the coagulopathy is not well defined

This study described haemostasis in a cohort of women with PPH to inform evidence-based treatment

Haemostatic impairment was uncommon during PPH but dilution of coagulation factors occurred with bleeds >3000 mL

A severe coagulopathy due to hyperfibrinolysis and hypodysfibrinogenaemia occurred in 1/1000 cases

Abstract

Background: Postpartum haemorrhage (PPH) may be exacerbated by haemostatic impairment. Information about PPH-associated coagulopathy is limited, often resulting in treatment strategies based on data derived from trauma studies.

Objectives: To investigate haemostatic changes associated with PPH.

Patients/methods: From a population of 11279 maternities, 518 (4.6%) women were recruited with PPH ≥1000 mL or placental abruption, amniotic fluid embolism (AFE) or concealed bleeding. Routine coagulation and viscoelastometric results were collated. Stored plasma samples were used to investigate women with bleeds >2000 mL or those at increased risk of coagulopathy defined as placenta abruption, AFE or need for blood components. Procoagulant factors were assayed and global haemostasis assessed using thrombin generation. Fibrinolysis was investigated with D-dimer and plasmin/antiplasmin (PAP) complexes. Dysfibrinogenaemia was assessed using the Clauss/antigen ratio.

Results: At 1000 mL blood loss Clauss fibrinogen was ≤2 g/L in 2.4% of women and 6/27 (22.2%) cases of abruption. Women with very large bleeds (>3000 mL) had evidence of a dilutional coagulopathy, although haemostatic impairment was uncommon. A subgroup of 12 women (1.06/1000 maternities) had a distinct coagulopathy characterised by massive fibrinolysis (PAP >40000 ng/mL), increased D-dimer, hypofibrinogenaemia, dysfibrinogenaemia, reduced factor V and factor VIII and increased activated protein C, termed acute obstetric coagulopathy (AOC). AOC was associated with fetal or neonatal death in 50% of cases and increased maternal morbidity.

Conclusions: Clinically significant haemostatic impairment is uncommon during PPH but a subgroup of women have a distinct and severe coagulopathy characterised by hyperfibrinolysis, low fibrinogen and dysfibrinogenaemia associated with poor fetal outcomes.

Key words

Coagulopathy, dysfibrinogenaemia, fibrinogen, fibrinolysis, postpartum haemorrhage

Introduction

Bleeding after childbirth (postpartum haemorrhage, PPH) causes about 57000 maternal deaths worldwide each year.[1] Obstetric bleeding may be exacerbated by haemostatic failure leading to massive PPH.[2] The characteristics of PPH-associated coagulopathy are poorly defined leading to some guidelines recommending the use of fixed-ratios of red blood cells (RBC), fresh frozen plasma (FFP) and platelets to resuscitate women,[3-7] based on data derived from major trauma studies in non-pregnant adults.[8 9]

Acute traumatic coagulopathy (ATC) is a complex dysfunction of haemostatic pathways driven by physical injury and shock. ATC is characterised by increase soluble thrombomodulin (sTM) which activates protein C, leading to inhibition of factor (F) Va and FVIIIa, whilst hyperfibrinolysis leads to clot beakdown and increased thrombin generation drives fibrin formation contributing to hypofibrinogenaemia. In addition, depletion and inhibition of coagulation factors due to dilution, hypothermia and acidosis during resuscitation exacerbate trauma-induced coagulopathy (TIC).[10-12] It is not known whether similar mechanisms occur during PPH.

PPH-associated coagulopathy has been described as a form of disseminated intravascular coagulation (DIC) where haemostatic changes are predominantly due to consumption of coagulation factors.[13] Management of DIC relies on infusion of FFP and platelets[14] but whether this is appropriate for PPH is debated.[2]

The haemostatic system at term differs from the non-pregnant state with increased procoagulant factors including fibrinogen, FVIII and VWF and reduced anticoagulants such as protein S.[15] This results in an increased capacity to generate thrombin[16] and substantially enhanced clot firmness measured by viscoelastometric haemostatic assays (VHAs). Due to these changes, any evolving coagulopathy during PPH will start from a different haemostatic baseline to those associated with trauma or DIC in the non-pregnant state.

It is established that fibrinogen falls to critically low levels earlier than other coagulation factors during PPH[17] with levels <2 g/L predictive of progression to massive PPH.[18-20] Guidelines therefore recommend maintaining Clauss fibrinogen above 2 g/L, or Fibtem >12mm, during PPH.[3-5 21] Clauss fibrinogen <2 g/L has been reported in 5% of cases of PPH of 1.5L, increasing to 17% in PPH $\geq$2.5L.[22 23] Prolongation of prothrombin time (PT) or activated partial thromboplastin time (aPTT) is rare during PPH until bleeds exceed 3L.[17] Formulaic treatment with FFP will therefore expose women with normal coagulation screens to unnecessary transfusion, whilst being inadequate to correct low fibrinogen.[24]

In contrast, some women develop severe peripartum coagulopathy which may be the main cause of haemorrhage. Classically this occurs in association with amniotic fluid embolism (AFE)[25] but may also be seen with placental abruption.[13 26] AFE-associated coagulopathy has been reported in small series and cases reports; these describe massive fibrinolysis, severe hypofibrinogenaemia, moderate thrombocytopenia and reduced FV and FVIII with normal levels of other procoagulant factors.[27-29] Recently, we reported a case of AFE with all these features and, in addition, an acute, acquired dysfibrinogenaemia.[30]

The mechanisms causing early fibrinogen depletion, relative to other coagulation factors, in PPH and the role of fibrinolysis are poorly understood. This study aimed to describe coagulation parameters during PPH and investigate PPH-associated coagulopathy to inform evidence-based treatment and improve outcomes for this potentially life-threatening complication of childbirth.

Methods

Study subjects

Women were recruited at 1000 mL measured blood loss, or earlier for clinical suspicion of placental abruption, AFE or concealed bleeding. All women giving birth had postpartum blood loss objectively measured, as opposed to estimated, by validated gravimetric and volumetric methods.[31] Consent to report clinical data and perform extended coagulation tests on stored samples was sought after the bleeding had stopped according to the ethical approval (REC16/WA/0282). Healthy term pregnant women (*n*=37) were recruited at the time of elective caesarean section to act as non-bleeding pregnant controls.

The causes of abnormal bleeding were recorded following review of the clinical notes and investigations such as placental histopathology. Many women had multiple causes of bleeding and the study team determined the primary cause of PPH in collaboration with the treating clinicians. The primary causes of bleeding were categorised as follows: uterine atony, surgical/trauma (including surgical bleeds and genital tract trauma), placental abruption, placenta accretia/praevia, retained products of conception (including retained placenta or membranes), AFE (diagnosis of the single case was made the day after delivery following notes review) and coagulopathy of unknown cause (no significant obstetric cause for bleeding was found and PPH was attributed to unexplained coagulopathy). Women were followed to discharge from hospital. Information about all women who gave birth during the study period was obtained from the local maternity unit database.

Laboratory testing

At recruitment, peripheral blood samples were taken for point-of-care testing, laboratory analysis, and extended testing. Point-of-care VHAs were performed using the Rotem Sigma (Werfen, Barcelona, Spain).[32] The PT, APTT and Clauss fibrinogen were analysed using an ACL TOP 700. Platelet poor plasma was obtained from citrated blood samples and stored at -80^{o}C for further analysis. Samples in a subgroup of women were taken into lithium heparin for measurement of circulating activated protein C (aPC). If bleeding continued, blood sampling was advised at every 500 mL blood loss or after infusion of coagulation products. Measured blood loss at the time of sampling was recorded where available.

Investigative testing of coagulation was performed on women with any of the following criteria; bleeds ≥2000 mL, placental abruption, AFE, treatment with fibrinogen concentrate or FFP or more than one blood sample taken during PPH. This group was selected to include women with the most severe bleeds and those most likely to be coagulopathic. Procoagulant factors FII, FV, FVII, FVIII, FIX, FX, FXI were measured by one stage clotting assay; and FXIII antigen and von Willebrand factor antigen (VWF:Ag) by latex enhanced immunoassay. The combined ability of coagulation factors to support haemostasis was assessed by thrombin generation measured on a Thrombinoscope (Thermo-Scientific) with peak thrombin and endogenous thrombin potential (ETP) reported. Dysfibrinogenaemia was investigated by antigenic fibrinogen, measured by ELISA, and the ratio of functional (Clauss) to total amount (ELISA) calculated. Fibrinolysis was assessed by measurement of D-dimers (reported as D-dimer units), plasminogen measured by chromogenic assay, plasmin/antiplasmin (PAP) complexes (ELISA) and maximum lysis quantified on Extem. The protein C pathway was investigated through measurement of protein C (chromogenic), aPC (ELISA) and soluble thrombomodulin (sTM) (ELISA). Detailed methods for laboratory analyses are provided in supplementary materials.

Clinical management

Women were managed according to the OBS Cymru Rotem algorithm.[33] Briefly, if bleeding was ongoing the algorithm prompted infusion of fibrinogen concentrate if the Fibtem A5 was <12 mm or Clauss fibrinogen <2 g/L, FFP infusion if the Extem CT was ≥75s or PT/aPTT were above the non-pregnant normal range, and platelet transfusion if the platelet count was <75 $x10^9$/L. Intravenous tranexamic acid (1g, repeated after 30 minutes if bleeding was ongoing) was given at 1000 mL blood loss or earlier if there was concern of concealed bleeding. Obstetric management was according to the All Wales PPH management guideline[34] based on guidelines from the Royal College of Obstetrics and Gynaecology.[4]

Data analysis and statistics

Descriptive analyses used median, interquartile range (IQR) and range for continuous variables and number and percent for categorial variables. Missing data were not imputed. Differences between groups were analysed using the Kruskal Wallis test with Bonferonni correction and Mann Whitney U or Chi square test (Excel and SPSS version 27).

Results

Between 6 May 2017 and 30 May 2019, there were 11279 maternities and 518 (4.6%) women with PPH were recruited. The median (IQR), range total blood loss was 1500 (1200-1800), 200-8500 mL. Table 1 shows the proportion of women eligible for the study that were recruited, and the proportion of women who had extended coagulation analyses performed (*n*=148). Of eligible women with ≥2000 mL blood loss, 101/106 (95.3%) were recruited and 83/101 (82.2%) had at least one extended blood test performed. The characteristics of recruited women are shown in Table 1.

Routine tests of coagulation and thromboelastometry at study entry

To investigate haemostatic changes early during the PPH, the blood samples taken at recruitment for routine laboratory and Rotem testing were analysed, dependent on the primary cause of bleeding (Table 2). At the time of first testing the median (IQR) blood loss was 1200 (1000-1400) mL.

Clauss fibrinogen: At study entry Clauss fibrinogen was ≤2 g/L in 11/449 (2.4%) cases, 6/27 (22%) abruptions and the cases of AFE and coagulopathy of unknown cause. Women with placental abruptions and praevia/accretia had lower Clauss fibrinogen levels on first measurement compared to pregnant term healthy controls (P<0.001). Results for Fibtem A5 were similar (Table 2).

Prothrombin time, activated partial thromboplastin time and Extem CT: At study entry the PT and aPTT were above the non-pregnant laboratory normal range in 6/449 (1.3%) and 1/449 (0.2%) cases, respectively and 9/483 (1.9%) women had an Extem CT ≥75 s. The median aPTT in women with PPH was 4s shorter than the lower end of the non-pregnant normal range.

Platelet count: At study entry 78/470 (16.6%) women had a platelet count below the normal range (150 $x10^9$/L) with 7/470 (1.5%) less than 75$x10^9$/L. Further description of the results is given in supplementary materials.

Summary of routine test results at study entry: These results demonstrate that reduced levels of coagulation factors, sufficient to prolong PT/aPTT, are very uncommon in women with PPH of about 1200 mL except for fibrinogen in the context of placental abruption and AFE. Despite this, it is known that some women develop early severe haemostatic failure at the time of childbirth. Studies in AFE

have highlighted that hyperfibrinolysis is an important component of these coagulopathies[27] and markers of this pathway were investigated.

Fibrinolysis during postpartum haemorrhage

PAP complexes were measured to investigate recent plasmin generation in 130 women with the largest bleeds (>2000 mL) and/or those most likely to have coagulopathy (abruption, AFE or receipt of blood products). These women had higher PAPs compared to the healthy term pregnant controls, median (IQR) 4760 (2531-16384) versus 1409 (1378-1481) (P<0.0001), non-pregnant normal range <512 ng/mL. D-dimers were higher in this group than healthy term pregnant controls median (IQR) 1659 (930-2990) versus 599 (410-866), P<0.0001, non-pregnant normal range <350 ng/mL consistent with activation of fibrinolysis during PPH.

Post hoc review of PAP distributions identified a distinct subgroup of 12 women who were significant outliers with PAPs >40000 ng/mL (Figures 1a, 1b). This group was defined as having acute obstetric coagulopathy (AOC). Women with AOC were compared to the other women who had had PAPs measured (*n*=118). Sequential samples had been taken from some women during the PPH and coagulation parameters for the sample associated with the highest PAP were analysed (Table 3).

Laboratory features of acute obstetric coagulopathy: D-dimers were >25-fold higher and platelets marginally lower in women with AOC compared to non-AOC. FV (Figure 1c) and FVIII were lower in AOC whilst other coagulation factors were similar between the groups, suggesting specific rather than generalised depletion of coagulation factors. The VWF/FVIII ratio was much higher in the AOC group because FVIII reduced whilst VWF:Ag increased. Despite falls in FV and FVIII, ETP and peak thrombin were similar between the groups.

Clauss fibrinogen was 49% lower in the AOC group, reduced to a median (IQR) of 2.1 (1.6-3.0) g/L (Figure 1d), a level associated with clinically significant haemostatic impairment in the context of PPH.[18 19 35] Similarly, reduced fibrinogen function was seen in the Fibtem assay with a fall of 42%. In contrast to Clauss fibrinogen, fibrinogen antigen levels were reduced by 20% in the AOC group and all had levels ≥2 g/L (Table 3). The Clauss/ELISA ratio was lower in the AOC group compared to non-AOC (Figure 3e) demonstrating that the reduced functional fibrinogen was caused by both a decreased amount of fibrinogen and acquired dysfibrinogenaemia. The non-AOC group had Clauss/ELISA ratios indistinguishable from healthy term pregnant controls and non-pregnant controls (Table 3). Plasminogen and FXIII levels were decreased in the AOC group.

Due to unavailability of sample tubes at the start of the study, aPC was only measured in 60 women with PPH. It increased from a median of 11 ng/ml in healthy term pregnant control women to 19.6 ng/ml in the non-AOC women with PPH (P<0.00001) (non-pregnant normal range 1.6-4.2 ng/ml). aPC was increased 4.5-fold in the AOC group compared to the non-AOC group (P=0.0007) (Table 3). Soluble thrombomodulin was higher in the healthy term pregnant controls than women with PPH (P<0.001) and there was no difference between the AOC and non-AOC groups.

Clinical features of acute obstetric coagulopathy: The clinical characteristics, maternal and neonatal outcomes of the 12 women who had evidence of AOC are shown in Table 4. AOC was associated with placental abruption in 5/12 cases and the case of AFE. The primary cause of bleeding for the other women was diverse.

At the time of the highest PAP, there was no difference in blood loss, lactate or shock index between the AOC and non-AOC groups (Table 3). Preeclampsia, smoking and lower gestation were more common in AOC but there was no association with clinical suspicion of sepsis (Table 4). AOC was

associated with maternal admission to the high dependency unit on delivery suite, increased RBC transfusion and infusion of fibrinogen concentrate. There was a markedly increased incidence of intra-uterine and neonatal deaths in the AOC group.

Tests of coagulation dependent on volume of postpartum haemorrhage

In addition to sample collection and testing at study entry, further samples were collected as bleed volume increased. Haemostasis results, dependent on measured blood loss at the time of sampling, are shown in Table 5 and median data are summarised in Figure 2. Women with AOC are excluded from these analyses.

Fibrinogen: Clauss fibrinogen was significantly lower at each bleed volume compared to pregnant term healthy controls (P<0.002). Data are influenced by the local treatment algorithm which prompted infusion of fibrinogen concentrate if the Fibtem A5 was <12 mm or Clauss fibrinogen <2 g/L leading to higher fibrinogen levels than if replacement had not happened, especially at larger bleed volumes.

Prothrombin time and activated partial thromboplastin time: The PT in women with PPH was marginally longer than the non-bleeding pregnant control group at all bleed volumes with a statistically significant increase in PT with larger bleed volumes (P<0.001). However, the number of women with PT above the non-pregnant normal range was small, 10/506 (2.0%). The aPTT was similar to healthy term pregnant controls until bleeds were >3000 mL. In bleeds >3000 mL, the aPTT was higher than the healthy term pregnant controls (P<0.005), however, only one woman had aPTT above the non-pregnant normal range. This woman had a bleed of 8500 mL due to undiagnosed placenta accrete. The aPTT became abnormal at 5700 mL blood loss, at which time she was given 2 units of FFP and the aPTT returned into the normal range.

Platelet count: The platelet count fell with increasing bleed volume and was statistically significantly lower than the healthy term pregnant controls for bleeds >2000 mL. These data are affected by the unit policy to transfuse platelets if they were below 75 $x10^9$/L. In total 6/506 (1.2%) women received platelets.

Individual coagulation factors and thrombin generation: Median values of FII, VII, VIII, IX and X in the healthy term pregnant women were higher than the mid-point of the laboratory reference range (table 3) whilst factor V and XI levels were not raised. These changes were associated with an increased peak thrombin and ETP and shortened PT and aPTT (table 3). FXIII was reduced in the healthy term pregnant controls to a median level of 54 IU/dL (laboratory normal range 64-136 IU/dL) (Table 3).

The median levels of individual coagulation factors and platelets varied according to bleed volume (Figure 3) (full data Table 5). Median levels of FII, V, VII, IX, X, XI and platelets fell linearly and in similar proportions as bleed volume increased. At bleed volumes >3000 mL, coagulation factors remained within the normal range except in 2 cases that had bleeds of 3000 and 8500 mL. Median FVIII did not reduce and tended to increase during PPH although this was not statistically significant (P=0.28). Factor VIII was within or above the population normal range in all women with PPH at all times. Median VWF:Ag increased in bleeds of 1000-1999 mL compared the non-bleeding pregnant controls (P <0.001). VWF:Ag was above 100 IU/dL in all cases of PPH at all times. In women with PPH, FXIII decreased in bleeds >2000 mL (P<0.0005) compared to healthy term pregnant controls and was a median of 27 IU/dL for bleeds >3000 mL (Table 5 and Figure 3).

The combined effect of procoagulant factors was assessed using thrombin generation. Peak thrombin was reduced in bleeds >2000 mL compared to healthy term pregnant controls (P <0.001) (Table 5) but remained increased compared to non-pregnant controls in bleeds up to 3000 mL (P <0.0001) and was similar for bleeds >3000 mL (P=0.4). The lowest peak thrombin in the PPH group was 161 nM, which is within the non-pregnant normal range. There were no statistically significant differences between the ETP of women of different bleed volumes or with healthy term pregnant controls. The ETP was >1000 nM/min and within the non-pregnant normal range in all cases of PPH. These data confirm adequate amounts of coagulation factors to support thrombin generation even at high bleed volumes.

D-dimer and plasmin/antiplasmin complexes: D-dimers were used as a potential marker of DIC in women without AOC. D-dimers were raised compared to healthy term pregnant controls for bleed volumes <999, 1000-1999 and 2000-2999 mL (P<0.005) but not for bleeds >3000 mL. There was no statistically significant difference in D-dimers between PPHs of different volumes. These results, combined with the linear decreases in individual coagulation factors and platelets, are compatible with loss due to bleeding and dilution being the main mechanism of coagulopathy associated with very large PPH.

PAPs were increased at all bleed volumes compared to healthy term pregnant controls (P<0.0001). There was a non-statistically significant trend for PAPs to increase as bleed volume increased.

Soluble thrombomodulin and activated protein C: sTM was decreased in women with PPH at all bleed volumes compared to healthy term pregnant controls (P<0.002) with no differences between bleed volumes. There was an increase in aPC for bleeds ≤999 mL and 1000-1999 mL compared to healthy term pregnant controls (P<0.001). The level of aPC for bleeds ≥2000 mL was similar to term controls.

Discussion

This study, performed in a large number of women with PPH, showed that abnormalities of coagulation are relatively uncommon in the context of treatment using our local monitoring and blood product replacement algorithm.[36 37] Most women maintain adequate haemostasis until large volumes of blood loss. In very large bleeds (>3000mL measured blood loss) evidence for an evolving dilutional coagulopathy was seen. We identified a small subgroup of women with PPH who had a distinct, severe coagulopathy characterised by hyperfibrinolysis and dysfibrinogenaemia.

Haemostatic impairment early during PPH (median 1200 mL blood loss) was uncommon, with only 2.4% having a Clauss fibrinogen <2 g/L and 1.3% an abnormal PT/aPTT, although reduced fibrinogen was more common in placental abruption, seen in 22% of cases, confirming findings from previous studies.[19 23 35] Thrombin generation in our study was increased at term similar to previous findings.[16] In this situation early replacement with FFP would not be expected to improve coagulation factor levels or haemostatic competence.[38 39]

In women with PPH who do not have AOC, the linear relationship between the fall in coagulation factors, except for FVIII, and platelets with bleed volume, suggests depletion and consumption of coagulation factors due to bleeding and clot formation and dilution due to resuscitation.[40 41] This is supported by the finding that, as coagulation factors declined, D-dimers did not increase suggesting that, in the non-AOC cases, DIC was not a prominent cause of the falling levels. The study did not measure thrombin/antithrombin complexes and so the amount of thrombin generated is not known. Individual coagulation factors remained within the non-pregnant normal range until bleeds were

larger than 3000 mL and even above that volume, most coagulation factors were normal. Factor V and XI were below the non-pregnant normal range in many cases but it is unlikely that this would have contributed to haemostatic impairment because thrombin generation remained normal. Normal thrombin generation potential, despite decreased levels of coagulation factors, is explained in part by stress-related increased FVIII. These findings suggest that FFP is unlikely to improve haemostasis except in very few cases and in our cohort only 3/518 (0.6%) women received FPP. The safety of withholding FFP based on VHAs has been shown previously.[42]

Lower levels of FXIII before labour, although remaining within the population normal range, are associated with an increased risk of PPH, independent of fibrinogen levels,[43 44] however, the FXIII level required for haemostasis during PPH is not known. Guidelines in inherited bleeding disorders recommend maintaining a level of above 20 IU/dL.[45] In bleeds >3L, all women had FXIII below the non-pregnant normal range although only one woman, total blood loss 8500 mL, had a level <20 IU/dL. The impact of these findings on *in vivo* haemostasis is unknown and investigation of the role of FXIII replacement, with cryoprecipitate or FXIII concentrate, during severe PPH is required. Fibrinogen concentrates contain different amounts of FXIII and their effect on *in vitro* haemostatic tests vary.[46] Cryoprecipitate reduced bleeding during PPH when given at the time of RBC transfusion, although it was challenging to give the cryoprecipitate in a timely fashion.[47]

Despite most women having normal haemostasis during PPH, it is recognised that a subgroup of women develop severe early haemostatic impairment that can precipitate massive PPH. In our study, 12/518 (2.3%) subjects (1.06/1000 maternities) had a specific coagulopathy, defined on the basis of massive hyperfibrinolysis (PAPs >40000 ng/ml), which we have called acute obstetric coagulopathy (AOC). Women with AOC formed a distinct group compared to other women with PPH in our study, with about a 30-fold increase in PAP levels and 25-fold increase in D-dimer confirming the presence of massive fibrinolysis. Ducloy-Bouthors investigated 144 women with PPH, without evidence of significant coagulopathy (mean fibrinogen above 3 g/L), and found average PAP levels of about 1000 ng/mL.[48] In major trauma studies, PAPs above 20000 ng/mL are used to define the most severe group.[49 50]

Women with AOC had a characteristic haemostatic profile with evidence for an acquired dysfibrinogenaemia that was not present in other women with PPH. This means that reduced Clauss fibrinogen and Fibtem A5 in the AOC group was due to both decreased absolute levels and impaired function. We have recently reported dysfibrinogenaemia in the context of severe PPH associated with AFE.[30] The mechanism for dysfibrinogenaemia in AOC is not known but may be related to high levels of fibrin degradation products interfering with fibrin polymerisation.[51]

AOC was associated with reduced FV, FVIII and FXIII but other procoagulant factors were preserved. Thrombin generation was normal or increased and there was a small decrease in platelet count. These haemostatic changes suggest a specific rather than generalised consumption of coagulation factors. Although FVIII was reduced in AOC, this would not be clinically significant because it remained above 100 IU/dL in all cases. The FVIII level was influenced by the rise in VWF and the relative depletion of FVIII was a more obvious when VWF/FVIII ratios were investigated.

FV and FVIII fell by similarly amounts, 43% and 39% respectively, suggesting that depletion may have been due to a shared mechanism. Depletion of FV and FVIII may have been caused by aPC as seen in acute traumatic coagulopathy.[10-12 52] Additionally, plasmin directly cleaves FV and FVIII[53 54] and this may have been a contributing mechanism given the massive excess of this enzyme in the AOC group. FXIII was about 17% lower in the AOC group compared to other women with PPH and non-bleeding healthy term women are known to have reduced FXIII levels.[55]

The coagulopathy in the AOC cohort is indistinguishable from that described in women with AFE where severe fibrinolysis and reduced fibrinogen, FV and FVIII and dysfibrinogenaemia are reported.[27-30] Yet in the cohort of 12 women with AOC described here only one was diagnosed with AFE, this diagnosis was made by the study team on notes review not the clinical team during the acute episode . The case of "coagulopathy of unknown cause" fits the UKOSS definition of AFE (https://www.npeu.ox.ac.uk/ukoss/current-surveillance/amf, accessed 16/Aug/2022) , although not the more strict criteria of Clark et al.[56] This diagnosis was not considered because the woman did not have circulatory collapse or hypoxia. AFE was reported in 3.3/100000 women with a 35% maternal mortality in a population-based study[57] and a literature review identified rates between 2.1-6.1/100000 depending on study methodology (https://www.npeu.ox.ac.uk/research/projects/66-afe-comparative-study, accessed 16/Aug/2022). These incidences are much lower than the 1.06/1000 women who developed AOC in our study, although subclinical AFE is a possible explanation. Five women with AOC had placental abruption and hypofibrinogenaemia is a known association. However, there were 26 other women with an abruption in our study with no evidence of AOC (average fibrinogen 4.1 g/l, PAPs 4733 ng/ml and D-dimer 2384 ng/ml) demonstrating that most abruptions are not affected. There were no fetal or neonatal deaths in these 26 cases. The remaining women with AOC had diverse causes of bleeding indicating that the coagulopathy can present in a variety of situations. These findings are clinically important because AOC cannot be predicted by the cause of bleeding, unless an AFE is diagnosed; and the only way to identify cases in an acute bleed is a low fibrinogen (Clauss or point-of-care) together with very high D-dimers (>20000 ng/ml) if this test is rapidly available.

AOC was associated with very poor fetal and neonatal outcomes when compared to other cases of PPH (50% (6/12) fetal/neonatal mortality vs 0.4% (2/506)). This is partly explained by the high incidence of placental abruption in the AOC group and poor outcomes are also reported in cases of pregnancy-related coagulopathy.[58] Whether the fetal and neonatal deaths were caused by or precipitated AOC requires further study. AOC was not associated with sepsis, a known risk factor for pregnancy-associated coagulopathy.[13] Despite having similar volumes of blood loss to other women with PPH, morbidity in women with AOC was increased and they were more likely to receive RBC transfusion, suggesting concealed bleeding, for example, into the uterine wall, and be admitted to high dependency care on the obstetric unit and intensive care although their shock index and lactate were similar to non-AOC cases. The clinical outcomes described in this paper should be considered in the context of the local PPH treatment algorithm which includes VHAs to detect low functional fibrinogen and guide replacement during cases of AOC. It is not possible to assess the likely outcomes in centres without access to early detection of hypofibrinogenaemia and there is the risk of progression of bleeding if fibrinogen replacement is delayed. The amount of fibrinogen needed to restore haemostasis in cases of AOC cannot be assessed from the data available if there is no access to VHAs, but higher doses than those required for dilutional coagulopathy may be necessary due to ongoing depletion and dysfibrinogenaemia.

The very high levels of PAP complexes and D-dimers show that excessive amounts of plasmin had been generated which cleaved crosslink fibrin. It is not known what precipitated the activation of plasminogen to plasmin or the activator responsible. A possible mechanism is that decreased levels of plasminogen activator inhibitors (PAI-1 or PAI-2) contributed although it is unlikely that the excessively high PAPs seen in the AOC group would have been caused through failure of inhibition alone.

In trauma, increased aPC is thought to be mediated by increased circulatory sTM associated with shock.[10-12] This does not seem to be the case in AOC because, whilst aPC was raised compared to

other women with PPH, sTM was not increased and bleed volume, shock index and lactate were similar between the AOC group and other women with PPH. The mechanism for the raised aPC in AOC requires further study.

The coagulopathy describe in AOC suggests that treatment should be with antifibrinolytics, such as tranexamic acid,[59] and fibrinogen replacement with cryoprecipitate or fibrinogen concentrate.[35 47] Despite the severe coagulopathy, the volume of bleeding between the AOC group and the non-AOC group was similar, suggesting that the treatment protocol used in our centre, which used early tranexamic acid and rapid identification and correction of low functional fibrinogen,[36 37] was effective in preventing bleed progression and there were no examples of AOC that were exacerbated by dilutional coagulopathy. Prospective studies are required to investigate this further.

This study has a number of limitations. Sample collection and storage performed outside of routine hours was challenging and not all women had samples available from every time point. In addition, extended tests of coagulation were performed on a subgroup of cases selected for bleed severity and likelihood of coagulopathy. It is not possible to exclude that cases of AOC were missed in the women who did not have extended testing but this is unlikely because women with low Clauss fibrinogen or Fibtem A5 were tested.

This was an exploratory study to investigate PPH-related coagulopathy. The women with AOC were categorised retrospectively because they were found to be outliers based on markers of hyperfibrinolysis. The coagulopathy described will need to be investigated further in prospective cohorts to establish whether the findings are reproducible. The mechanisms that precipitated very high levels of PAPs and raised aPC were not determined and require further study. We were unable to analyse aPC in the complete cohort due to the initial lack of appropriate blood bottles; therefore the aPC data are limited and should be interpreted with caution.

In conclusion, in a large cohort of women with PPH, that included the large majority of women in our institution with bleeds >2000mL, very few develop haemostatic impairment early during the bleed. Women without AOC have a linear reduction in coagulation factors and platelets with increasing bleed volume, compatible with consumption into clots and dilution, although no woman had reduced thrombin generation.

We identified a cohort of women (1.06/1000 maternities) who had a distinct and severe early coagulopathy, that we termed acute obstetric coagulopathy. This coagulopathy was indistinguishable from that previously reported for AFE. The AOC group were not identifiable by clinical presentation alone and recognition and treatment of this coagulopathy depends on availability of timely coagulation results. The local treatment protocol of tranexamic acid infusion and maintaining fibrinogen >2g/L appeared to be effective in limiting progression of PPH associated with AOC. The implications of reduced FXIII levels in pregnant women at term and during PPH are unclear and require further investigation. Overall, our results suggest that empirical fixed-ratio treatment of PPH with early FFP is unlikely to improve coagulation in most women with PPH and will lead to large numbers receiving unnecessary blood products and under-treatment of women with established coagulopathy. Emphasis for correction of PPH-related coagulopathy should be on administration of anti-fibrinolytic agents and replacement of fibrinogen. How and when to test for fibrinogen during a PPH, and the optimal source for replacement is, requires further study.

Authorship details

The manuscript has been read and approved by all authors.

LdeL was the study principal investigator, led the ethical approval, designed the study, collated data, interpreted data and co-wrote the first draft of the manuscript.

PVJ designed the study, led the laboratory investigations, interpreted data and co-wrote the first draft of the manuscript.

SFB designed the study, collated data, interpreted data and co-wrote the first draft of the manuscript.

NJM interpreted data and critically reviewed the manuscript.

JF performed the thrombin generation assays, collated data, interpreted data and critically reviewed the manuscript.

PMB developed and provided materials for the aPC test, interpreted data and critically reviewed the manuscript.

DJ co-led the consenting of women, developed and maintained the study database, collated data, interpreted data and critically reviewed the manuscript.

AR co-led the consenting of women, collated data, interpreted data and critically reviewed the manuscript.

LC collated data, interpreted data and critically reviewed the manuscript.

TR collated data, interpreted data and critically reviewed the manuscript.

VF collated data, interpreted data and critically reviewed the manuscript.

REC designed the study, collated data, interpreted data and co-wrote the first draft of the manuscript.

PWC designed the study, interpreted data and co-wrote the first draft of the manuscript.

Acknowledgements

We thank the women who had been through the trauma associated with postpartum haemorrhage who agreed to take part in the study . Some had suffered tragic personal loss and all wished for their experience to contribute towards increasing understanding of postpartum haemorrhage and to improving safety in childbirth, and without whom this study would not have been possible.

Staff on labour ward and in the Haematology and Blood Transfusion laboratory who were busy providing emergency care to women yet still managed the research samples.

Samantha Drew, Genevieve McCluskey, Martin Davies and Maria Watkins for laboratory testing.

Research Midwives: Sian Jones, Maryanne Bray, Emily Brace, Emma Davies

Anaesthetic research trainees and fellows: Zain Amir, Bronwen Price, Chloe Wilson, Steffan Merrix, Christopher Coomber, Simran Sharma, Thomas Kitchen, Bethan Morris, Lucy French, William Packer,

Catherine Griffiths, Nicola Boyer, Rob Sparrow, Hannah Johnson-Hughes, Chloe Wilson, Rowenna Morris-Clarke, Isra Hassan, Mike Adamson

Obstetric Trainee: Cerys Scarr

Research nurse: Doria Barboncheilli

Research Officer: Rachel Purser

Funding

The study was funded by grants from The National Institute of Academic Anaesthesia, Obstetric Anaesthetist Association and Haemonectics Corporation. None of the grant giving bodies played any role in study design, data collection, interpretation of results or decision to publish. The study received support from Health Care Research Wales and Cardiff and Vale University Health Board Research and Development office. PVJ was supported by Health Care Research Wales NHS Research Time Award (2016-19) and Sir Geraint Evans Cardiovascular Research Fund Award (2019).

Conflicts of interest

LdeL has received research support Haemonentics.

SFB has received research support and paid consultancies from Werfen and Haemonentics.

NJM has received research support and paid consultancies from Alveron Pharma, LFB Group and STAGO.

REC has received research support and paid consultancies from CSL Behring, Werfen and Haemonentics.

PWC has received research support and paid consultancies from CSL Behring, Werfen and Haemonentics.

None of the other authors have conflicts to declare.

Tables

Table 1. Characteristics of women giving birth and recruited to the study

	All maternities N=11279	**Women recruited N=518**	**Extended tests performed N=148**	**P value All women recruited vs extended tests group**
Age years Med (IQR)	30 [26-34]	31.5 [28-35]	32.5 [28-35]	0.31
Body mass index Med (IQR)	26 [23-30]	26 [23-31]	25 [23-30]	0.44
Gravida N (%)	ND	2 [1-3]	2 [1-3]	0.06
Parity N (%)	ND	1 [0-2]	1 [0-2]	0.33
Induction of labour N (%)	ND	181 (35%)	55 (37%)	0.62
Augmented labour N (%)	ND	144 (28%)	36 (24%)	0.7
Mode of birth				
Unassisted vaginal N (%)	7056 (62.6)	165 (32%)	39 (26%)	0.2
Instrumental vaginal N (%)	1469 (13.0)	130 (25%)	38 (26%)	0.89
Elective CS N (%)	1397 (12.4)	80 (15%)	24 (16%)	0.81
Non-elective CS N (%)	1370 (12.1)	143 (28%)	47 (32%)	0.32
Primary cause of PPH[1]				
Abruption N (%)	ND	31 (6%)	20 (13.5%)	0.002
Atony N (%)	ND	71 (13.7%)	16 (10.8%)	0.36
Surgical/trauma N (%)	ND	334 (64.5%)	76 (51.4%)	0.004
Placental accretia/praevia N (%)	ND	26/518 (5%)	13 (8.8%)	0.09
Retained products of conception N (%)	ND	54 (10.4%)	21 (14.2 %)	0.2
Amniotic fluid embolus N (%)	ND	1 (0.2%)	1 (0.7%)	0.34
Coagulopathy of unknown cause N (%)	ND	1 (0.2%)	1 (0.7%)	0.34
Volume of PPH				
Total measured blood loss mL Med (IQR)	350 (200-550)	1500 (1205-1800)	2000 (1585-2300)	<0.001
Not known N (%)	503 (4.5)	0	0	NA
0-999 mL N (%)	9783 (86.7)	23 (4.4)	7 (4.7)	NA
1000-1999 mL N (%)	887 (7.9)	394 (76.1)	58 (39.2)	NA
2000-2999 mL N (%)	89 (0.8)	86 (16.6)	70 (47.3)	NA

3000-3999 mL N (%)	13 (0.1)	11 (2.1)	9 (6.1)	NA
≥4000 ml N (%)	4 (0.04)	4 (0.8)	4 (2.7)	NA
Transfusion				
Any red blood cell transfusion N (%)	ND	133 (26%)	72 (49%)	<0.001
Fibrinogen concentrate N (%)	ND	19 (3.7%)	18 (12.2%)	<0.001
FFP transfusion N (%)	ND	3 (0.6%)	3 (2%)	0.1
Platelet transfusion N (%)	ND	7 (1.4%)	4 (2.7%)	0.26

Legend: 1. Primary cause of bleeding as assessed by the study team, many women had multiple causes of bleeding with atony contributing to 290 cases and surgical/trauma to 384. NA is not applicable and ND is no data available.

Table 2. Routine tests of coagulation and thromboelastometry performed at study entry

	Non-pregnant healthy control	**Pregnant term healthy controls N=37**	**All PPH N=518**	**Placental abruption N=31**	**Atony N=71**	**Surgical or Trauma N=334**	**RPOC N=54**	**Placenta accretia or praevia N=26**	**AFE N=1**	**Coagulopathy unknown cause N=1**
Measured blood loss at first sample (mL) Median (IQR), range	NA	None	1200 (1000-1400) 70-3000	600 (400-1100) 70-1300	1300 (1085-1500) 350-2200	1200 (1013-1460) 100-3000	1100 (1000-1300)	1000 (950-1290) 300-2320	2000	ND
Hb at first sample (g/L) Median (IQR), range	115-165	121 (116-126) 104-136	109 (100-119) 67-150	108 (100.5-128) 67-150	113 (101.5-122.5) 72-150	109 (99-118) 70-149	114 (105-124) 86-141	102.5 (94-109) 67-119	135	115
Clauss fibrinogen g/L Median (IQR), range N (%) ≤2 g/L	2-4[1]	5.0 (4.4-5.6) 3.5-7.9 0/37 (0%)	4.4 (3.7-5.2) 0.4-10.1 11/449 (2.4)	3.7 (2.7-4.5) 1.3-6.0 6/27 (22.2)	4.6 (3.7-5.7) 2.3/7.3 0/60	4.5 (3.8-5.3) 1.8-8.3 2/292 (0.7)	4.8 (4.1-5.1) 1.8-10.1 1/44 (2.3)	3.8 (3.6-4.6) 2.5-6.1 0/25 (0)	1.7 - - 1/1 (100)	1.4 - - 1/1 (100)
PT (sec) Median (IQR), range N (%) above NR	9-13[1]	10.4 (9.9-10.6) 9.1-11.5	10.8 (10.4-11.3) 8.6-20.1 6/449 (1.3)	10.6 (10.2-11.4) 9.5-13.3 1/27 (3.7)	10.8 (10.5-11.3) 9.5-12.1 0/60 (0)	10.8 (10.4-11.3) 8.6-14.7 4/292 (1.4)	10.9 (10.5-11.3) 9.4-12.7 0/44 (0)	10.7 (10.4-11.3) 9.9-12.2 0/25 (0)	11.8 - - 0/1 (0)	20.1 - - 1/1 (100)
aPTT (sec) Median (IQR), range N (%) above NR	28-38.5[1]	25.1 (23.4-26.0) 20-29.8	24.1 (22-25.9) 20-46.1 1/449 (0.2)	24.4 (22.7-26.2) 20-29.5 0/27 (0)	23.9 (20.0-25.6) 20-29.4 0/60 (0)	24.2 (22.1-26.1) 20-37.7 0/292 (0)	23.1 (20.3-25.2) 20-46.1 1/44 (2.3)	23.9 (21.2-25.6) 20-27.6 0/25 (0)	25.5 - - 0/1(0)	31.3 - - 0/1 (0)
Platelet count (x10^9/L) Median (IQR), range N (%) below NR	150-400[1]	230 (181-279) 101-419 4/36 (11.1)	200 (163-237) 19-438 78/470 (16.6)	197 (173-262) 19-371 3/27 (11.1)	203 (166-238) 53-414 10/66 (15.1)	200 (161-235) 53-438 53/302 (17.5)	194 (173-253) 116-399 5/49 (10.2)	225 (142-193) 63-303 7/24 (29.2)	203 - - 0/1 (0)	180 - - 0/1 (0)
Fibtem A5 (mm) Median (IQR), range N (%) <12 mm	ND	23 (21-26) 6.0-33	20 (17-24) 0-41 16/483 (3.3)	17 (12-22) 4-30 7/28 (25)	20 (16/23) 9-39 1/63 (1.6)	20 (17-24) 7-41 6/317 (1.9)	22 (19-25) 12-37 0/49 (0)	18 (15-22) 12-31 0/24 (0)	6 - - 1/1 (100)	0 - - 1/1 (100)
Extem CT (sec) Median (IQR), range N (%) ≥75 s	ND	56 (52-62) 43-80	54 (50-59) 51-323 9/483 (1.9)	61 (50-71) 47-97 4/28 (14.3)	54 (52-58) 46-90 1/63 (1.6)	53 (50-57) 41-77 1/317 (0.3)	56 (52-60) 45-79 1/49 (2.0)	54 (50-59) 46-68 0/24 (0)	94 - - 1/1 (100)	323 - - 1/1 (100)
Extem Maximum lysis N (%) >15%	11 (2.5-17) 0-22	5 (2.0-8.5) 0-15 0 (0%)	5 (1-9) 0-25 4/481 (0.8)	2.5 (0.5-6.5) 0-11 0/28 (0)	5 (1-8) 0-15 0/63 (0)	6 (1-9) 0-25 3/316 (0.9)	5.5 (1-9) 0-12 0/48 (0)	5 (0-9) 0-19 1/23 (4.3)	4 - - 0/1 (0)	0 - - 0/1 (0)

Legend: Laboratory normal and reference ranges (NR)[1]. ND is data not available, NA is not applicable, RPOC: retained products of conception and includes retained placenta and/or membranes. Blood loss in the placental abruption group is lower because cases were recruited at the time of diagnosis rather than at 1000 mL and it is likely that some bleeding was concealed.

Table 3. Haemostatic variables and indices of shock in acute obstetric coagulopathy

	Non-pregnant healthy controls (laboratory normal range or reference ranges)	**Non-bleeding term pregnant controls N=37**	**Non-acute obstetric coagulopathy group Median (IQR) Range N=118**	**Acute obstetric coagulopathy group Median (IQR) Range N=12**	**P Non-AOC vs AOC**
Blood loss when samples taken (mL) Median (IQR), range	NA	0	1500 (1125-2000) 875-5700	1350 (1085-2300) 595-5500	0.9
Shock index when samples taken Median (IQR), range	NA	NA	0.85 (0.73-1.0) 0.4-1.75	0.81 (0.60-0.91) 0.51-0.94	0.35
Lactate when samples take (mmol/L) Median (IQR), range	0.5-1.6	ND	2.3 (1.9-2.8) 1.1-7.5	2.1 (1.5-2.7) 0.9-3.5	0.14
D-dimer (ng/mL) Median (IQR), range	<350	599 (410-866) 239-2226	1702 (915-2726) 240-17438	43915 (14283-58085) 10607-64145	<0.0001
Plasminogen (IU/dL) Median (IQR), range	80-120	ND	103 (91-120) 37-173	82 (70-100) 54-124	<0.05
Platelets ($x10^9$/L) Median (IQR), range	150-400	230 (181-279) 101-419	187 (150-251) 19-435	149 (109-172) 98-184	<0.01
aPTT (sec) Median (IQR), range	27-38.5	25.1 (23.4-26.0) 20-29.8	24.5 (22.2-26.4) 20-63.3	27.2 (24.8-29.6) 22.9-34.8	<0.05
Clauss fibrinogen (g/L) Median (IQR), range	2.8 (2.5-3.3) 1.8-4.9	5.9 (4.4-5.6) 3.5-7.9	4.1 (3.4-5.0) 1.6-9.0	2.1 (1.6-3.0) 0.4-4.2	<0.0001
Fibtem A5 (mm) Median (IQR), range	ND	23 (21-26) 6.0-33	20 (16-24) 7-37	11.5 (8-14) 0-21	<0.0001
Fibrinogen ELISA (g/L) Median (IQR), range	3.5 (3.2-4.1) 2.0-7.2	6.4 (5.8-7.4) 4.7-8.5	5.0 (4.3-5.9) 1.8-10.1	4.0 (3-4.5) 2-5.6	<0.005
Fibrinogen Clauss/ELISA ratio Median (IQR), range	0.80 (0.76-0.87) 0.66-0.96	0.76 (0.72-0.84) 0.57-10.4	0.83 (0.72-0.92) 0.47-1.20	0.57 (0.48-0.68) 0.21-0.91	<0.0001
Factor II (IU/dL)	50-200	145 (132-155) 92-172	109 (97-123) 32-177	96 (71-119) 61-161	0.10
Factor V (IU/dL) Median (IQR), range	50-200	108 (99-118) 74-222	88 (73-105) 16-174	50 (32-77) 16-102	<0.0005
Factor VII (IU/dL) Median (IQR), range	50-200	188 (173-204) 123-232	141 (118-161) 48-239	169 (119-187) 98-217	0.14
Factor VIII (IU/dL) Median (IQR), range	50-200	205 (162-263) 108-496	262 (179-351) 86-908	159 (59-315) 50-440	<0.05
VWF:Ag (IU/dL) Median (IQR), range	50-200	283 (266-309) 163-587	357 (273-442) 78-950	439 (299-711) 259-1783	0.08
VWF/FVIII ratio Median (IQR), range	ND	1.38 (1.05-1.88) 0.59-2.58	1.29 (1.05-1.69) 0.28-5.60	3.57 (2.26-5.29) 1.34-8.54	<0.0001
Factor IX (IU/dL)	50-150	173	158	122	<0.05

Median (IQR), range		(159-188) 74-267	(133-178) 56-275	(111-158) 68-277	
Factor X (IU/dL) Median (IQR), range	50-150	159 (142-179) 122-283	115 (99-132) 31-203	98 (79-117) 62-183	0.12
Factor XI (IU/dL) Median (IQR), range	50-150	108 (92-120) 59-163	82 (66-96) 19-195	78 (58-96) 36-172	0.66
Factor XIII (IU/dL)	64-136	54 (43-81) 22-98	54 (43-65) 15-136	45 (34-51) 19-60	<0.01
Peak thrombin generation (nM) Median (IQR), range	238 (206-295) 110-408	397 (347-446) 194-532	382 (330-426) 138-561	355 (302-355) 264-673	0.68
Endogenous thrombin potential (nM/min) Median (IQR), range	1527 (1372-1750) 624-2644	2067 (1944-2367) 1273-3008	2030 (1738-2381) 1015-3435	2089 (1886-2379) 1224-3808	0.58
Soluble thrombomodulin (pg/mL) Median (IQR), range	16.5 (15.0-20.0) 12.0-25.3	42.7 (34.0-45.2) 26.5-53.8	29.6 (23.1-36.3) 8.7-117.3	29.7 (27.2-46.0) 19.2-48.4	0.32
Protein C (IU/dL) Median (IQR), range	70-151	134 (118-150) 84-196	92 (790-110) 23-172	82 (65-106) 48-171	0.35
Highest activated protein C (IU/dL) during bleeding Median (IQR), range	2.4 (2.0-3.3) 1.6-4.2[1] N=70	11 (9.7-12.3) 3.1-17.5 N=21	19.6 (15.2-34.9) 2.4-172 N=54	88.7 (46.4-121) 38-993 N=6	0.0007

Legend: [1]The normal range for aPC was performed in a different laboratory to the study samples and so may not be directly comparable. AOC is acute obstetric coagulopathy. Results in the AOC and non-AOC groups correspond to the time of the highest plasmin/antiplasmin complex except for activated protein C where the highest recorded level is reported. ND is data not available, NA is not applicable.

Table 4. Clinical features of mother and neonate in the acute obstetric coagulopathy group

	All non-acute obstetric coagulopathy N=506	Non-acute obstetric coagulopathy with PAPs performed N=118	Acute obstetric coagulopathy N=12	P All non-acute obstetric coagulopathy **vs** acute obstetric coagulopathy
Age (years)	31.5 (28-35) 16-49	32 (28-35) 16-45	30.5 (27-35) 21-45	0.8
BMI: Median (IQR, range)	26 (23-31) 16-55	25 (23-30) 17-53	24 (24-29) 20-41	0.5
Parity: Median (IQR, range)	1 (0-2) 0-10	1 (0-2) 0-5	2 (1-2) 0-5	0.045
Gestation (weeks) Median (IQR, range)	40 (38-41) 17-42	40 (37-41) 17-42	36.5 (29.7-39) 20-40	0.0002
Induction of labour: N (%)	180 (35.6)	49 (41.5)	1 (8.3)	0.05
Multiple birth: N (%)	24 (4.7)	8 (6.8)	1 (8.3)	0.6
Smoker: N (%)	35 (6.9)	9 (7.6)	3 (25)	0.02
Preeclampsia: N (%)	41 (8.1)	11 (9.3)	4 (33.3)	0.01
Mode of birth				
Vaginal unassisted: N (%)	160 (31.6)	30 (25.4)	5 (41.7)	0.5
Instrumental vaginal: N (%)	129 (25.5)	33 (28.0)	1 (8.3)	0.2
Elective CS: N (%)	78 (15.4)	16 (13.6)	2 (16.7)	0.9
Non elective CS: N(%)	139 (27.5)	39 (33.1)	4 (33.3)	0.7
Cause of postpartum haemorrhage				
Placental abruption: N(%)	26 (5.1)	15 (12.7)	5 (41.7)	0.008
AFE: N(%)	0 (0)	0 (0)	1 (8.3)	NA
Coagulopathy: N(%)	0 (0)	0 (0)	1 (8.3)	NA
Atony: N(%)	70 (13.8)	9 (7.6)	1 (8.3)	0.93
Surgical: N(%)	333 (65.8)	67 (56.8)	1 (8.3)	0.001
Praevia/accrete: N(%)	24 (4.7)	9 (7.6)	2 (16.6)	0.28
RPOC: N(%)	53 (10.5)	18 (15.3)	1 (8.3)	0.52
Total blood loss (mL) Median (IQR, range)	1500 (1222-1800) 200-8500	2000 (1600-2378), 200-8500	1525 (1050-2650), 200-5500	0.9

Red blood cell transfusion (units) Median (IQR, range)	0 (0-0) 0-19	0 (0-2) 0-16	1.5 (1-2.5) 0-9	<0.0001
Fibrinogen concentrate infusion: N (%)	9/506 (1.8%)	8/118 (6.8)	10/12 (83.3)	<0.0001
Sepsis pathway: N (%)	72 (14)	23 (19)	1 (8)	0.6
HDU admission N (%)	304/485 (62.7)	104 (88.1)	12 (100)	0.03
ITU admission: N (%)	1 (0.2)	0 (0)	1 (8.3)	<0.0001
Hysterectomy: N (%)	4 (0.8)	3 (2.5)	0	NA
Intra-uterine death: N (%)	1 (0.2)	1 (0.8)	5 (41.7)	<0.0001
Neonatal death: N (%)	1 (0.2)	0 (0)	1 (8.3)	<0.0001
Intra-uterine or neonatal death: N (%)	2 (0.4)	1 (0.8)	6 (50)	<0.0001

Legend: HDU is high dependency unit on delivery suite, ITU is intensive case unit, BMI is body mass index, NA is not applicable.

Table 5. Coagulation tests by blood loss at the time of testing

	0 mL[1]	**<999 mL**	**1000-1999 mL**	**2000-2999 mL**	**>3000 mL**
Clauss fibrinogen (g/L) Median (IQR), range	5.1 (4.6- 5.6) 3.8-7.2	4.1 (3.7-4.7) 2.4-7.6 N=59	4.5 (3.7-5.3) 1.8-10.1 N=367	3.6 (2.8-4.6) 1.6-9.0 N=55	2.5 (2.1-3.3) 1.7-5.1 N=14
PT (sec) Median (IQR), range	10.4 (9.9-10.6) 9.1-11.5	10.7 (10.3-11.2) 9.7-12.6 N=59	10.8 (10.4-11.3) 8.6-14.7 N=367	11.1 (10.6-12.0) 9.5-13.6 N=55	11.6 (11.0-14.1) 10.2-18.2 N=14
aPTT (sec) Median (IQR), range	25.1 (23.4-26.0) 20-29.8	24.5 (20.9-26.4) 20-31.4 N=59	24.1 (22.2-25.9) 20.0-37.7 N=367	24.5 (22.4-28.2) 20.0-34.4 N=55	27.6 (25.3-35.0) 20.0-63.3 N=14
Platelet count ($x10^9$/L) Median (IQR), range	230 (181-279) 101-419	195 (162-235) 63-419 N=64	201 (161-244) 53-438 N=378	172 (138-206) 85-336 N=59	127 (92-162) 67-217 N=14
D-dimer (ng/mL) Median (IQR), range	599 (410-866) 239-2226	1248 (705-1938) 192-3883 N=18	1739 (876-2895) 240-17438 N=104	1768 (841-3021) 286-20176 N=45	1019 (560-2121) 265-7436 N=11
PAP (ng/mL) Median (IQR), range	1409 (1378-1481) 1284-2119 N=20	2304 (2014-3102) 2304-16643 N=15	3197 (2140-7844) 1136-97424 N=95	3396 (2483-5869) 1176-23975 N=41	4103 (1899-7415) 1657-9160 N=10
Factor II (IU/dL) Median (IQR), range	145 (132-155) 92-172	127 (113-136) 88-142 N=18	113 (97-124) 47-177 N=104	97 (83-109) 43-145 N=45	79 (46-94) 32-119 N=11
Factor V (IU/dL) Median (IQR), range	107 (99-118) 74-222	99 (84-123) 62-189 N=18	93 (78-104) 29-174 N=104	71 (55-98) 34-144 N=45	51 (23-73) 16-137 N=11
Factor VII (IU/dL) Median (IQR), range	188 (173-204) 123-232	148 (127-168) 88-300 N=18	147 (119-166) 59-232 N=104	122 (101-156) 52-196 N=45	99 (89-123) 46-153 N=11
Factor VIII (IU/dL) Median (IQR), range	205 (162-263) 108-496	226 (172-354) 57-551 N=18	253 (169-329) 50-908 N=104	248 (147-358) 53-559 N=45	180 (106-302) 57-492
Factor IX (IU/dL) Median (IQR), range	173 (159-188) 74-267	156 (125-191) 102-246 N=18	155 (136-178) 42-155 N=104	135 (111-166) 74-247 N=45	94 (72-163) 56-190 N=11
Factor X (IU/dL) Median (IQR), range	159 (142-179) 122-283	127 (106-137) 86-191 N=18	117 (103-132) 44-203 N=104	99 (80-112) 42-166 N=45	68 (47-89) 31-113 N=11
Factor XI (IU/dL) Median (IQR), range	107 (92-120) 59-163	85 (66-97) 42-112 N=18	79 (64-95) 24-153 N=104	68 (50-88) 27-126	51 (31-79) 19-131 N=11
Factor XIII (IU/dL) Median (IQR), range	54 (43-81) 22-98	52 (37-64) 30-92 N=20	52 (44-56) 15-136 N=104	44 (31-53) 14-135 N=45	27 (25-38) 15-50 N=11
VWF:Ag (IU/dL)	283	306	362	316	258

Median (IQR), range	(266-309) 163-587	(271-380) 168-646 N=18	(273-439) 78-950 N=104	(258-450) 162-640 N=45	(174-286) 140-388 N=11
Peak thrombin (nM) Median (IQR), range	397 (347-446) 194-532	390 (347-427) 265-482 N=18	382 (329-426) 117-538 N=107	340 (301-376) 193-561 N=38	289 (203-326) 161-350 N=9
Endogenous thrombin potential (nM/min) Median (IQR), range	2067 (1944-2367) 1273-3008	2083 (1834-2580) 1501-3433 N=18	2011 (1738-2381) 1025-3435 N=107	1932 (1676-2223) 1015-3006 N=38	1814 (1667-2029) 1431-2250 N=9
Soluble thrombomodulin (pg/mL) Median (IQR), range	42.7 (34.0-45.2) 26.5-53.8	32.2 (25.4-36.5) 15.1-43.6 N=15	29.5 (22.4-36.7) 14.6-117.3 N=94	25.3 (22.6-31.1) 11-107.1 N=41	24.3 (12.5-37.7) 7.0-46.3 N=10
Activated protein C (IU/dL) Median (IQR), range	11 (9.7-12.3) 3.1-17.5 N=21	33.7 (12-36.6) 8.8-69.2 N=7	19 (15.2-28.4) 7.3-136 N=35	17.9 (11.4-27) 2-52.2 N=15	13 (8.6-15.9) 8.5-16.5 N=5

Legend. The 12 women with acute obstetric coagulopathy have been excluded from this table. Samples taken before elective caesarean section[1]. An individual woman may contribute data to multiple bleed volume brackets. Data for bleeds <1000 mL are not representative because woman were recruited before 1000 mL for clinical concern of an abruption, AFE or concealed bleeding. The results are not adjusted for administration of fibrinogen concentrate (*n*=9) or FFP (*n*=3), this means that the data will tend to overestimate the fibrinogen level because the treatment algorithm aimed to maintain the fibrinogen >2 g/L if bleeding was ongoing. FFP replacement will have had minimal effect. N relates to number of samples not number of women.

Figure 1. Plasmin/antiplasmin complexes, factor V and dysfibrinogenaemia in women with postpartum haemorrhage

Legend. The highest PAP levels in 130 women with severe postpartum haemorrhage are shown. A distinct group of outliers with PAPs >40000 ng/mL defined the group of women labelled as acute obstetric coagulopathy (AOC) in the further analyses (a and b). The levels of FV, Clauss fibrinogen and Clauss to antigenic fibriogen ratio are shown in the 118 non AOC and 12 AOC women are shown (c,d,e). Women with AOC had reduced FV and an acquired dysfibrinogenaemia.

Figure 2. Coagulation factor levels and platelet number dependent of volume of bleed

Legend: Lines represent the median values of each parameter for women at the specified bleed volume. Full data are given in Table 5. Women with acute obstetric coagulopathy have been excluded. The “no bleeding” group are samples taken before elective caesarean section. An individual woman may contribute data to multiple bleed volume brackets. Data for bleeds <1000 mL are not representative because woman were recruited before 1000 mL for clinical suspicion of a placental abruption, AFE or concealed bleeding. The results are not adjusted for administration of fibrinogen concentrate (*n*=9) or FFP (*n*=3), this means that the data will overestimate the fibrinogen level because the treatment algorithm aimed to maintain the fibrinogen >2 g/L if bleeding was ongoing. FFP replacement will have minimal effect. Y axes are for coagulation factors on the left and platelets on the right.

References

1. Say L, Chou D, Gemmill A, et al. Global causes of maternal death: A WHO systematic analysis. *The Lancet Global Health* 2014;2(6):e323-e33.
2. Collis R, Collins P. Haemostatic management of obstetric haemorrhage. *Anaesthesia* 2015;70(Supple 1):78-86.
3. Collins P, Kadir R, Thachil J. Management of coagulopathy associated with postpartum haemorrhage: guidance from the SSC of ISTH. *Journal of Thrombosis and Haemostasis* 2016;14:205-10.
4. Mavrides E, Allard S, Chandraharan E, et al. Prevention and management of postpartum haemorrhage. *British Journal of Obstetrics and Gynaecology: An International Journal of Obstetrics and Gynaecology* 2016;124:e106-e49.
5. Escobar MF, Nassar AH, Theron G, et al. FIGO recommendations on the management of postpartum hemorrhage 2022. *International Journal of Gynecology and Obstetrics* 2022;157(S1):3-50.
6. WHO. WHO recommendations for the prevention and treatment of postpartum haemorrhage 2012 [Available from: https://www.who.int/reproductivehealth/publications/maternal_perinatal_health/9789241548502/en/.
7. Muñoz M, Stensballe J, Ducloy-Bouthors AS, et al. Patient blood management in obstetrics: Prevention and treatment of postpartum haemorrhage. A NATA consensus statement: A multidisciplinary consensus statement. *Blood Transfusion* 2019;17(2):112-36.
8. Holcomb JB, Tilley BC, Baraniuk S, et al. Transfusion of plasma, platelets, and red blood cells in a 1:1:1 vs a 1:1:2 ratio and mortality in patients with severe trauma: The PROPPR randomized clinical trial. *JAMA - Journal of the American Medical Association* 2015;313(5):471-82.
9. Spahn DR, Bouillon B, Cerny V, et al. The European guideline on management of major bleeding and coagulopathy following trauma: fifth edition. *Critical care (London, England)* 2019;23(1):98. doi: 10.1186/s13054-019-2347-3
10. Curry NS, Davenport R. Transfusion strategies for major haemorrhage in trauma. *British Journal of Haematology* 2019;184(4):508-23.
11. Moore HB, Gando S, Iba T, et al. Defining trauma-induced coagulopathy with respect to future implications for patient management: Communication from the SSC of the ISTH. *Journal of Thrombosis and Haemostasis* 2020;18(3):740-47.
12. Davenport R, Brohi K. Causes of trauma-induced coagulopathy. *Current opinions in Anesthesiology* 2016;29:212-19.
13. Erez O. Disseminated intravascular coagulation in pregnancy: New insights. *Thrombosis Update* 2022;6 doi: 10.1016/j.tru.2021.100083.
14. Wada H, Thachil J, Di Nisio M, et al. Guidance for diagnosis and treatment of disseminated intravascular coagulation from harmonization of the recommendations from three guidelines. *Journal of Thrombosis and Haemostasis* 2013;11(4):761-67.
15. Allard S, Green L, Hunt BJ. How we manage the haematological aspects of major obstetric haemorrhage. *British Journal of Haematology* 2014;164(2):177-88.
16. McLean KC, Bernstein IM, Brummel-Ziedins KE. Tissue factor-dependent thrombin generation across pregnancy. *American Journal of Obstetrics and Gynecology* 2012;207(2):135.e1-35.e6.
17. De Lloyd L, Bovington R, Kaye A, et al. Standard haemostatic tests following major obstetric haemorrhage. *International Journal of Obstetric Anesthesia* 2011;20(2):135-41.
18. Charbit B, Mandelbrot L, Samain E, et al. The decrease of fibrinogen is an early predictor of the severity of postpartum hemorrhage. *Journal of Thrombosis and Haemostasis* 2007;5(2):266-73.
19. Collins PW, Lilley G, Bruynseels D, et al. Fibrin-based clot formation as an early and rapid biomarker for progression of postpartum hemorrhage: a prospective study. *Blood* 2014;124(11):1727-36.

20. Cortet M, Deneux-Tharaux C, Dupont C, et al. Association between fibrinogen level and severity of postpartum haemorrhage: Secondary analysis of a prospective trial. *British Journal of Anaesthesia* 2012;108(6):984-89.
21. Curry NS, Davenport R, Pavord S, et al. The use of viscoelastic haemostatic assays in the management of major bleeding: A British Society for Haematology Guideline. *British Journal of Haematology* 2018;182(6):789-806.
22. Bell SF, Collis RE, Bailey C, et al. The incidence, aetiology, and coagulation management of massive postpartum haemorrhage: a two-year national prospective cohort study. *International Journal of Obstetric Anesthesia* 2021;47, 102983.
23. Bell SF, Collis RE, Collins PW. Comparison of haematological indices and transfusion management in severe and massive postpartum haemorrhage: analysis of a two-year national prospective observational study. *International Journal of Obstetric Anesthesia* 2022;50, 103547
24. Collins PW, Bell SF, Lloyd d, et al. Management of postpartum haemorrhage: from research into practice, a narrative review of the literature and the Cardiff experience. *International Journal of Obstetric Anesthesia* 2018;37:106-17.
25. Amniotic fluid embolism syndrome: analysis of the Unites States International Registry. American Journal of Obstetrics and Gynecology MFM; 2020;2:100083.
26. McNamara H, Mallaiah S, Barclay P, et al. Coagulopathy and placental abruption: Changing management with ROTEM-guided fibrinogen concentrate therapy. *International Journal of Obstetric Anesthesia* 2015;24(2):174-90.
27. Schröder L, Hellmund A, Gembruch U, et al. Amniotic fluid embolism-associated coagulopathy: a single-center observational study. *Archives of Gynecology and Obstetrics* 2020;301(4):923-29.
28. Shen F, Wang L, Yang W, et al. From appearance to essence: 10 years review of atypical amniotic fluid embolism. *Archives of Gynecology and Obstetrics* 2016;293(2):329-34.
29. Matsunaga S, Takai Y, Nakamura E, et al. The clinical efficacy of fibrinogen concentrate in massive obstetric haemorrhage with hypofibrinogenaemia. Scientific Reports, 2017;7:46749.
30. Oliver C, Freyer J, Murdoch M, et al. A description of the coagulopathy characteristics in amniotic fluid embolism: a case report. *International Journal of Obstetric Anesthesia* 2022; 51:103573
31. Powell E, James D, Collis R, et al. Introduction of standardised, cumulative quantitative measurement of blood loss into routine maternity care *The Journal of Maternal-Fetal & Neonatal Medicine* 2022;35:1491-1497.
32. Bell SF, Roberts TCD, Freyer Martins Pereira J, et al. The sensitivity and specificity of rotational thromboelastometry (ROTEM) to detect coagulopathy during moderate and severe postpartum haemorrhage: a prospective observational study. *International Journal of Obstetric Anesthesia* 2022;49:103238.
33. Available from: https://phw.nhs.wales/services-and-teams/improvement-cymru/our-work/maternity-cymru/obs-cymru/obstetric-bleeding-strategy-cymru/rotem-point-of-care-testing/.
34. Maternity Network Wales. Prevention and management of postpartum haemorrhage 2017 [Available from: http://www.wisdom.wales.nhs.uk/sitesplus/documents/1183/All%20Wales%20Prevention%20of%20Postpartum%20Haemorrhage%20%281.4%29_2018.pdf.
35. Collins PW, Cannings-John R, Bruynseels D, et al. Viscoelastometric-guided early fibrinogen concentrate replacement during postpartum haemorrhage: OBS2, a double-blind randomized controlled trial. *British Journal of Anaesthesia* 2017;119(3):411-21.
36. Bell SF, Collis RE, Pallmann P, et al. Reduction in massive postpartum haemorrhage and red blood cell transfusion during a national quality improvement project, Obstetric Bleeding Strategy for Wales, OBS Cymru: an observational study. *BMC Pregnancy and Childbirth* 2021;21(1):377.

37. Bell SF, Kitchen T, M. J, et al. Designing and Implementing an All Wales Postpartum Haemorrhage Quality Improvement Project: OBS Cymru (The Obstetric Bleeding Strategy for Wales) *BMJ Quality* 2020;9:e000854.
38. Chowdhury P, Saayman AG, Paulus U, et al. Efficacy of standard dose and 30 ml/kg fresh frozen plasma in correcting laboratory parameters of haemostasis in critically ill patients. *British Journal of Haematology* 2004;125(1):69-73.
39. Collins PW, Solomon C, Sutor K, et al. Theoretical modelling of fibrinogen supplementation with therapeutic plasma, cryoprecipitate, or fibrinogen concentrate. *British Journal of Anaesthesia* 2014;113(4):585-95.
40. Bolliger D, Görlinger K, Tanaka KA. Pathophysiology and treatment of coagulopathy in massive hemorrhage and hemodilution. *Anesthesiology* 2010;113(5):1205-19.
41. Hiippala ST, Myllyla GJ, Vahtera EM. Hemostatic factors and replacement of major blood loss with plasma-poor red cell concentrates. *Anesthesia and Analgesia* 1995;81(2):360-65.
42. Collins PW, Cannings-John R, Bruynseels D, et al. Viscoelastometry guided fresh frozen plasma infusion for postpartum haemorrhage: OBS2, an observational study. *British Journal of Anaesthesia* 2017;119(3):422-34.
43. Haslinger C, Korte W, Hothorn T, et al. The impact of prepartum factor XIII activity on postpartum blood loss. *Journal of Thrombosis and Haemostasis* 2020;18(6):1310-19.
44. Karlsson O, Jeppsson A, Hellgren M. Factor XIII activity at onset of labour and association with postpartum haemorrhage: an exploratory post-hoc study. *International Journal of Obstetric Anesthesia* 2021;47:103174.
45. Pavord S, Rayment R, Cumming T, Lester W, Chalmers E, Myers B, Maybury H, Tower C, Kadir R, on behalf of the Royal Colleg of Obstetrics and Gynaecology. . Management of Inherited Bleeding Disorders in Pregnancy. *BJOG : an international journal of obstetrics and gynaecology* 2017;124:e193-e263.
46. Whyte CS, Rastogi A, Ferguson E, et al. The Efficacy of Fibrinogen Concentrates in Relation to Cryoprecipitate in Restoring Clot Integrity and Stability against Lysis. *International Journal of Molecular Sciences* 2022;23(6):23062944.
47. Green L, Daru J, Gonzalez Carreras FJ, et al. Early cryoprecipitate transfusion versus standard care in severe postpartum haemorrhage: a pilot cluster-randomised trial. *Anaesthesia* 2022;77(2):175-84.
48. Ducloy-Bouthors AS, Duhamel A, Kipnis E, et al. Postpartum haemorrhage related early increase in D-dimers is inhibited by tranexamic acid: Haemostasis parameters of a randomized controlled open labelled trial. *British Journal of Anaesthesia* 2016;116(5):641-48.
49. Cardenas JC, Matijevic N, Baer LA, et al. Elevated tissue plasminogen activator and reduced plasminogen activator inhibitor promote hyperfibrinolysis in trauma patients. *Shock* 2014;41(6):514-21.
50. Raza I, Davenport R, Rourke C, et al. The incidence and magnitude of fibrinolytic activation in trauma patients. *Journal of Thrombosis and Haemostasis* 2013;11(2):307-14.
51. Latallo ZS, Budzyński AZ, Lipiński B, et al. Inhibition of thrombin and of fibrin polymerization, two activities derived from plasmin-digested fibrinogen [28]. *Nature* 1964;203(4950):1184-85.
52. Keyloun JW, Le TD, Orfeo T, et al. Assessing Factor V Antigen and Degradation Products in Burn and Trauma Patients. *Journal of Surgical Research* 2022;274:169-77.
53. Lee CD, Mann KG. Activation/inactivation of human factor V by plasmin. *Blood* 1989;73(1):185-90.
54. Pasquini R, Hershgold EJ. Effects of plasmin on human factor VIII (AHF). *Blood* 1973;41(1):105-11.
55. Sharief LT, Lawrie AS, Mackie IJ, et al. Changes in factor XIII level during pregnancy. *Haemophilia* 2014;20(2):e144-e48.
56. Clark SL, Romero R, Dildy GA, et al. Proposed diagnostic criteria for the case definition of amniotic fluid embolism in research studies. *American Journal of Obstetrics and Gynecology* 2016;215(4):408-12.

57. Roberts CL, Algert CS, Knight M, et al. Amniotic fluid embolism in an Australian population-based cohort. *BJOG: An International Journal of Obstetrics and Gynaecology* 2010;117(11):1417-21.
58. Sohn CH, Kim SR, Kim YJ, et al. Disseminated Intravascular Coagulation in Emergency Department Patients with Primary Postpartum Hemorrhage. *Shock* 2017;48(3):329-32.
59. Shakur H, Roberts I, Fawole B, et al. Effect of early tranexamic acid administration on mortality, hysterectomy, and other morbidities in women with post-partum haemorrhage (WOMAN): an international, randomised, double-blind, placebo-controlled trial. *The Lancet* 2017;389(10084):2105-16.